Personal Copy:

Vol. 1, No. 2 April 1958

PAKISTAN JOURNAL OF SCIENTIFIC AND INDUSTRIAL RESEARCH

Published by

PAKISTAN COUNCIL OF SCIENTIFIC AND INDUSTRIAL RESEARCH
KARACHI

Personal Copy.

PAKISTAN JOURNAL OF SCIENTIFIC AND INDUSTRIAL RESEARCH

Vol. 1, No. 2 April 1958

CONTENTS

Some Reactions of Gramine.....S.K. Qureshi, K. Rasheed, A.K. Mekeri and George Hahn 101

Prevention of Malaria Transmission with Single Dose of Pyrimethamine (Daraprim).....Colonel M.K. Afridi and Abdul Rahim 107

Evaluation of Fungicidal Properties of Lignin Acetic Acid. Part II.—Studies on Treated Jute Seeds in Infested Soil.....M.I. Ali, Q.A. Ahmed and M.H. Khundkar 113

Studies in the Relationship Between Viscosity and Molecular Structure. Part I.—Temperature Dependence of the Viscosity of Certain Typical Mineral Oils.....Mazhar Mahmood Qurashi 116

Preliminary Structural Study of the Magnetic Fraction of Steam-Treated Makerwal Coal.....S. Jamal Quadir and Sadrul Hasan Rizvi 128

Preliminary Ultra-Violet Absorption Study of the Halogen Acid Salts of Alkaloids of the Conessine Series.....A.J. Hamdani, Noorul Haq Khan, and the late S. Mehdi Ahsan 132

An Investigation of the Correction for Peripheral Heat-Loss from Thick Samples in Lees and Chorlton's Disc Method of Determining Thermal Conductivity.....A.K.M. Ahsanullah and Mazhar Mahmood Qurashi 136

Chemical Investigation of Neem-Seeds.....A.M. Ahsan and George Hahn 146

Determination of Optimum Conditions for Cooking and Treating Intact Fish Slices with Special Reference to Bone Softening..... M. Qudrat-i-Khuda, S. Fazl-i-Rubbi, H. Rahman and N.A. Khan 151

Studies on Shark Liver Oil. Part II.—Recovery of Vitamin 'A' by Different Digestion Methods.....S. Maqsood Ali, S. Abdul Haque and S. Mahdihassan 156

Cobalt Content of East Pakistan Fishes.....Mohammad Abul Hussain Sharif and Kamaluddin Ahmed 158

Estimation of Vitamin B12 by Colorimetric Determination of Cobalt.....Mohammad Abul Hussain Sharif 160

The Pupa and Larva of Eublemma Scitula.....S. Mahdihassan 164

Chaksine. Part II.—Pyrolysis of Chaksine.....Salimuzzaman Siddiqui, Ahmad Kamal, George Hahn and (in part) Vishwa Nath Sharma 167

Chaksine. Part III. .....Ahmad Kamal, Mohammad Ayub Bokhari, Louis Fernandez and George Hahn 168

Non-Technical Notes

COTTONSEED OIL-BASED PAINTS AND OTHER COATING COMPOSITIONS..... 173

SHARK LIVER OIL AND VITAMIN A CONCENTRATES..... 175

SUBSTITUTE FOR BIDI LEAVES..... 176

PREPARATION OF MIXED BILE SALTS AND ACIDS FROM BILE..... 177

INSULATING TAPES..... 178

PETROL GAS PLANT..... 179

Notes and News..... 180

Book Notice..... 183

Patent News..... 184

Pakistan Journal of Scientific and Industrial Research

EDITORIAL BOARD

Salimuzzaman Siddiqui, D. Phil. Nat.
Director, Scientific and Industrial Research
(Ex-Officio Chairman)

Col. M. K. Afridi, M. D., D.T.M. & H.
Vice-Chancellor, University of Peshawar,
& Director, North Regional Laboratories,
Pakistan Council of Scientific and
Industrial Research, Peshawar

Ali Ahmad, Ph. D., D.I.C.
Operative Director, Chemical Industries,
Pakistan Industrial Development
Corporaton, Karachi

Taskhir Ahmad B.Sc. (Agr.), Ph.D.
Director, Department of Plant Protection,
Karachi

M. Innas Ali, M.Sc,M.E.E., Ph.D.
Head of the Department of Physics,
University of Dacca, Dacca

M. O. Ghani, M.Sc. Ph.D.
Agricultural Development Commissioner,
East Pakistan, Dacca

Mazhar-ul Haque, M.D.
Professor and Head of the Department of
Pharmacology, Dow Medical College,
Karachi

S. Hedayatullah, M.Sc., Ph.D.
Liaison Officer for East Pakistan,
Ministry of Agriculture, Govt. of Pakistan,
Head Quarter, Dacca

Q. M. Hussain, M.A., Ph.D.
Dean, Faculty of Science,
University of Dacca, Dacca

Karimullah, D. Phil. Nat., Ph.D.
Director, University Institute of Chemistry,
Lahore

M. Qudrat-i-Khuda, D.Sc., D.I.C., M.Sc.
Director, East Regional Laboratories,
Pakistan Council of Scientific and Industrial
Research, Dacca

M. Raziuddin Siddiqi,
M.A., Ph.D., D.Sc., Member, Atomic
Energy Commission, Karachi

M. M. Qurashi,
B.A. (Hons.) M.Sc., Ph.D., A. Inst. P.
Senior Research Officer,
Central Laboratories, Pakistan Council of
Scientific and Industrial Research, Karachi
(Editor & Ex-Officio Secretary)

M. A. Haleem, M.Sc., M.A., LL.B.
Patent Officer, Pakistan Council of
Scientific and Industrial Research, Karachi
(Associate Editor)

Papers should be widely-spaced, single-sided type-scripts in English, wide margins being provided to facilitate editorial instructions to the printers. Publication will be speeded if a duplicate copy accompanies the manuscript. The submission of a manuscript will be taken to imply that the material has not been previously published in any language.

Line drawings for the manuscript should preferably be 6″, 9″, 12″ wide, and be planned for reduction to the width of a single column. The drawing may be on white drawing paper or tracing cloth, and the lines should be black and reasonably broad to ensure clear reproduction. Photographic reproductions (of drawings) that conform to the above dimensions will also be acceptable. Photo-micrographic and other pictures should be glossy prints with a maximum black and white contrast. They should be rectangular in shape and should not be marred by heavy writing or paper clips, staples, etc. All drawings and photographs should carry on the back the figure number and the author's name. Figure captions should be typed and collected on a separate sheet at the end of the manuscript.

Tables should have suitable captions at the top, and should be carefully laid out with brief and self-explanatory column headings, economy being exercised in arranging the tables. Mathematical equations should be carefully printed by hand without any over-writing. Greek and other unusual symbols should be identified in the margin, thus "Greek Alpha."

References should be numbered consecutively and arranged thus at the end of manuscripts :

1. Q.M. Ahmad, Phys. Rev., **41**, 852 (1932).

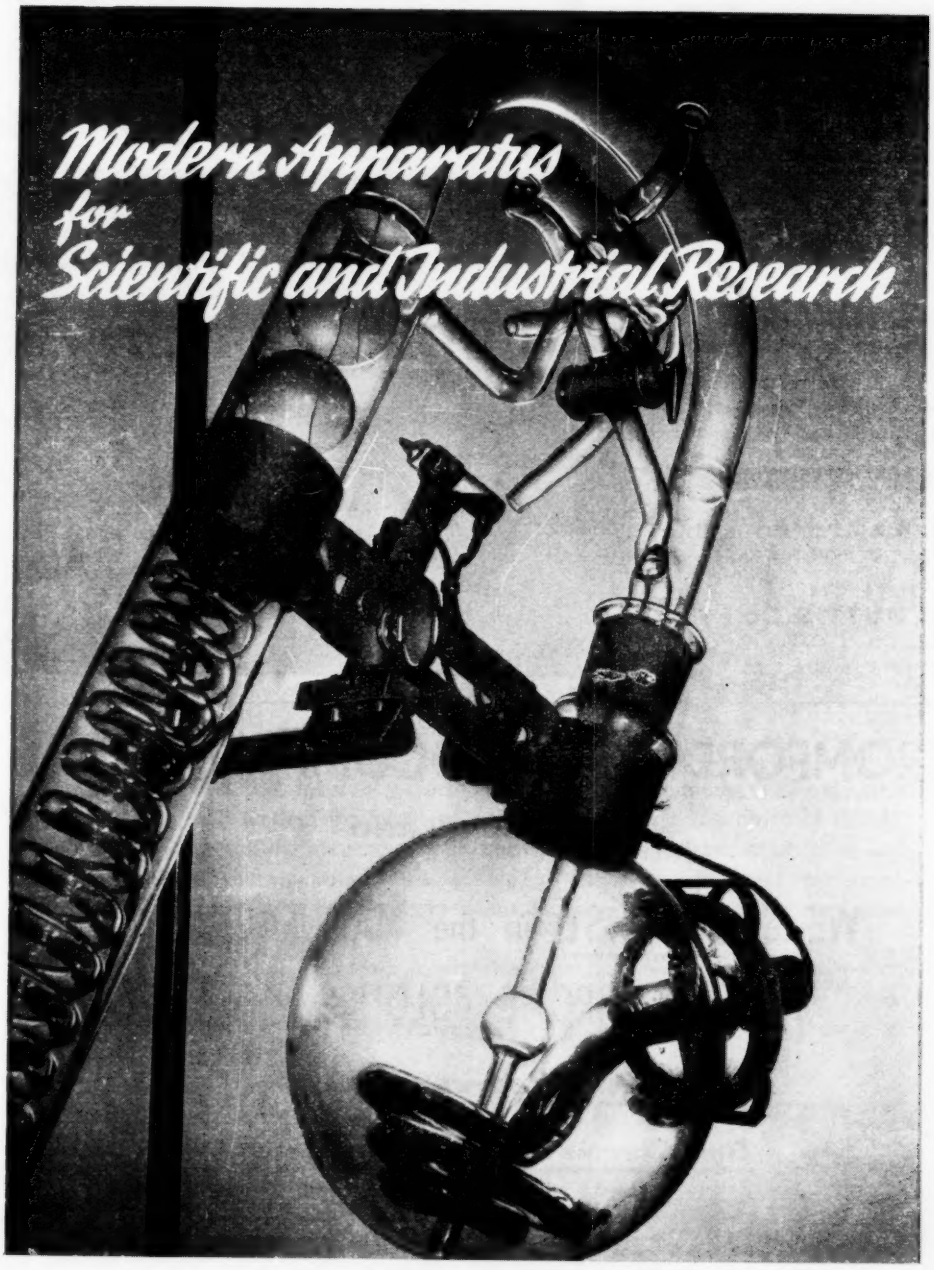
2. H. Lamb, *Hydrodynamics* (Cambridge University Press, Cambridge, England, 1940), sixth edition, pp. 573, 645.

Work included in the manuscript but done by the author in other laboratories should be clearly acknowledged. Data in the form of figures or tables quoted from sources other than the manuscript should refer to the sources in the captions of the figure or table.

All proofs will be sent to the author and should be returned promptly, extensive alterations being avoided. Orders for reprints should accompany the corrected proofs.

All manuscripts and enquiries should be addressed to the Associate Editor, Pakistan Council of Scientific and Industrial Research, No. 3/4/D/IV, Nazimabad, Karachi.

Orders should be sent to the Associate Editor. Annual subscription Rs. 15/- post free. Single copies, Rs. 4/- post free.



KARL KOLB

SCIENTIFIC-TECHNICAL-SUPPLIES

FRANKFURT-MAIN I . GERMANY

MOLECULAR WEIGHTS

.. A quick new way

Gallenkamp Ebulliometer

HEITLER PATTERN

* Determinations in 10 minutes
* Accuracy 1% with 0.2 g samples
* Rapidly reaches equilibrium (< 3 minutes)
* Thermistor sensing for rapid response and high sensitivity
* Borosilicate glass parts
* Additional equipment not required
* Simple to use
* Quickly saves its own cost in time gained.

Designed in co-operation with Mr. C. Heitler of Northampton College of Advanced Technology (London).

A. GALLENKAMP & CO. LTD.,
SUN STREET, LONDON, E. C. 2.

Appointed Agents :

TAQI BUTT & Co.
3 Mall Mansions, P. O. Box 62,
The Mall, Lahore.

CROMFORD COLOUR COMPANY LTD.

MATLOCK, ENGLAND

WE SPECIALISE in the MANUFACTURE

of CHROME COLOURS

Lead Chrome Yellows and Greens

Zinc Chrome Yellows and Greens

Barium Chromate

Representative in Pakistan

Moh'd Siddiq Abdul Majid
SOUTH NAPIER ROAD, KARACHI-2

A new Symbol of Quality

Responsible laboratory workers in many countries recognise a B.D.H. label as a symbol of quality. To strengthen this confidence still further, the labels of up to 1,500 items in the range of B.D.H. laboratory reagents will in future carry details of minimum assay and of the maximum limits allowed of the more significant impurities. New labels, grey-green in colour, have been designed to accommodate this information.

Reagents carrying these specifications will gradually replace the corresponding materials under existing labels and it is hoped that the new development will be cordially welcomed by users of B.D.H. chemicals.

B.D.H. LABORATORY CHEMICALS

THE BRITISH DRUG HOUSES LTD. B.D.H. LABORATORY CHEMICALS GROUP **POOLE ENGLAND**

Distributors:

West Pakistan — **Bliss & Company,** P.O. Box No. 7113, Karachi, Sadar. Telephone: 51463

East Pakistan — **Shafi & Company Ltd.** 1 Wiseghat Road, Dacca. Telephone: 2629

PRECIOUS METALS

IN ANY FORM AND FOR EVERY PURPOSE
REWORKING AND REFINING
CRUCIBLES, DISHES, ELECTRODES
SALTS, CATALYSTS AND SEMI-FINISHED MATERIALS

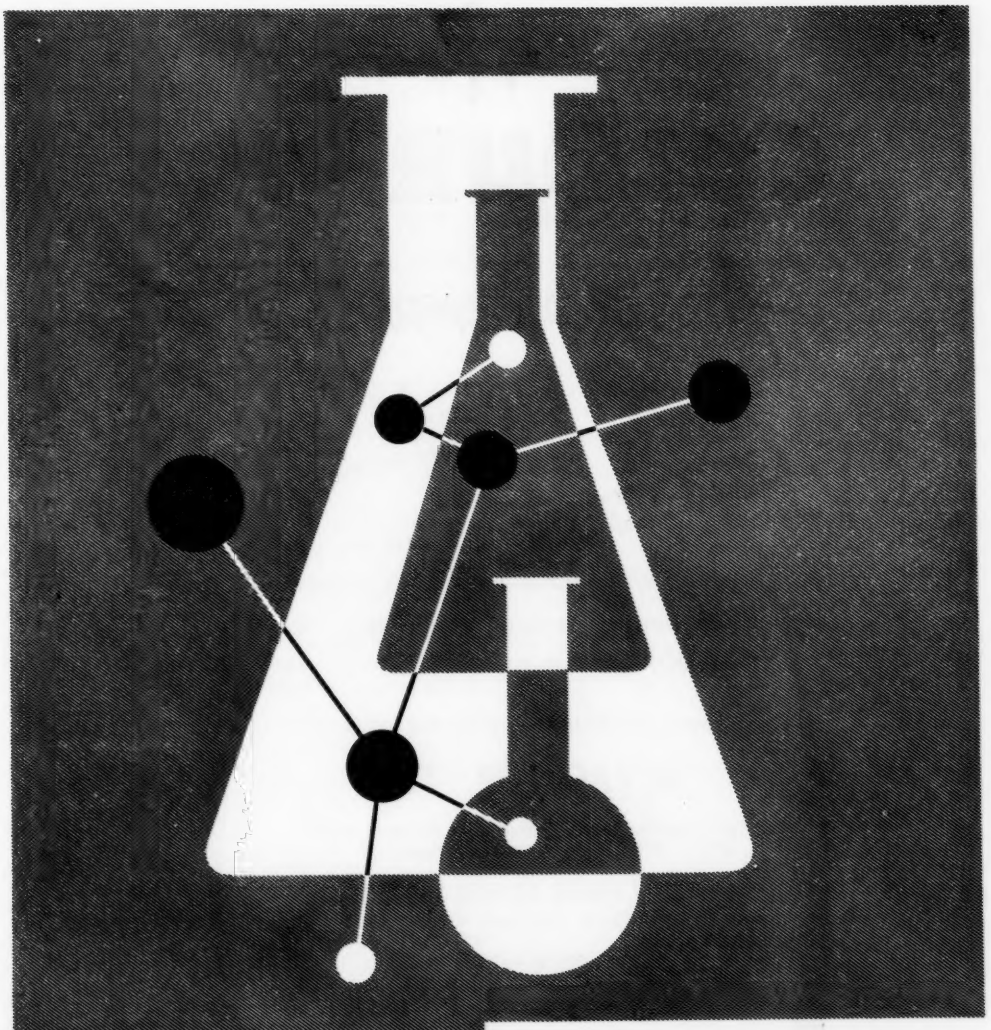
LABORATORY HEAT TREATING EQUIPMENT, ELECTRICALLY HEATED SUCH AS:

TUBE FURNACES UP TO 1800°C
MUFFLE FURNACES UP TO 1000°C
CHAMBER FURNACES UP TO 1100°C AND 1350°C, RESP.
DRYING OVENS OF CLYINDRICAL AND RECTANGULAR SHAPE
VACUUM DRYING OVENS
VACUUM DESICCATORS
INCUBATORS OF CYLINDRICAL AND RECTANGULAR SHAPE
WATER, OIL, AND AIR BATHS
CRUCIBLE FURNACES
HOT PLATES
COMBUSTION FURNACES FOR MACRO AND MICRO ELEMENTARY ANALYSES.

QUARTZGLASS

PURE TRANSPARENT QUARTZGLASS
TRANSLUCENT QUARTZGLASS
QUARTZGLASS FOR OPTICAL PURPOSES
LABORATORY UTENSILS OF QUARTZ, ELECTRICALLY HEATED
WATER-DISTILLATION APPARATUSES
INFRARED RADIATORS

W. C. HERAEUS GMBH HANAU (Western Germany)



CHEMICALS

Merck

E. Merck

DARMSTADT · GERMANY

Sole Agents in West Pakistan
ABDUL SATTAR SONS & CO.
47, NOOR CHAMBERS
ROBSON ROAD, KARACHI

Sole Agents in East Pakistan
CAPCO AZIZ LIMITED
344, JUBILEE ROAD
CHITTAGONG

A COLOURFUL Announcement

GEVACOLOR

We have pleasure to announce that arrangements have now been completed for processing, printing and enlarging of Gevacolor negatives in Pakistan.

Capture the colour magic of life and nature with

GEVACOLOR FILM

and return your exposed film to your dealer who will immediately forward it to :

RAINBOW PHOTO COLOUR LABORATORY,
QAMAR HOUSE, BUNDER ROAD, KARACHI

for processing and printing on

GEVACOLOR PAPER

The true to life colour prints will bring back to you the wonderful memories in glorious colours

THE SIGN OF PROGRESS

PHOTO GEVAERT CINE

Allied Photographics (Pakistan) Ltd.

Karachi — Lahore — Chittagong

STRONACHS

Pakistan Journal

of

Scientific and Industrial Research

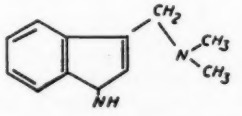
Vol. I, No. 2 April 1958

SOME REACTIONS OF GRAMINE

S. K. Qureshi, K. Rasheed, A.K. Mekeri and George Hahn

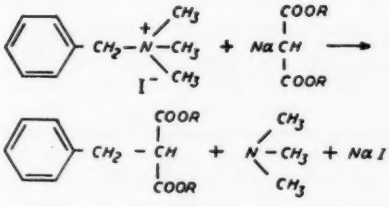
Central Laboratories, Pakistan Council of Scientific and Industrial Research, Karachi

Gramine (I) was first isolated out of chlorophyll deficient mutants of barley by v. Euler and Hellstroem[1] in 1932 and was soon found to be identical with the alkaloid donaxine out of *Arundo Donax* (*Gramineae*).[2] Kuhn and Stein[3] found a synthesis using the Mannich reaction which, applied on indole with formaldehyde and dimethylamine in acetic acid solution gives quantitatively pure gramine.



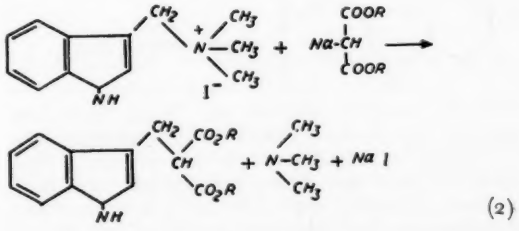
I

It gained considerable importance when Snyder[4] in 1944 used it for an excellent synthesis of tryptophane. This achievement was based on studies of the reaction of quaternary ammonium salts containing the benzyl moiety. According to equation 1 they were found to act as alkylating agents yielding benzyl malonate (65%).



(1)

By using graminemethyl iodide, Snyder found that this salt behaves in the same manner furnishing skatyl malonate with even better yields (85%) according to equation (2).

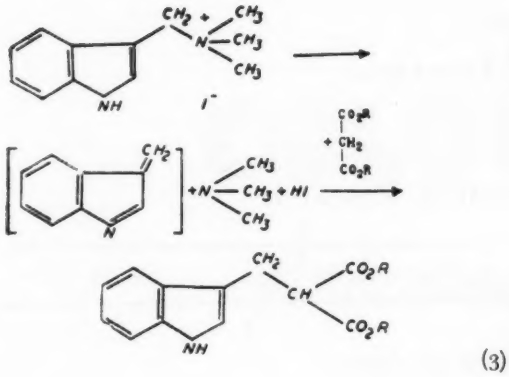


(2)

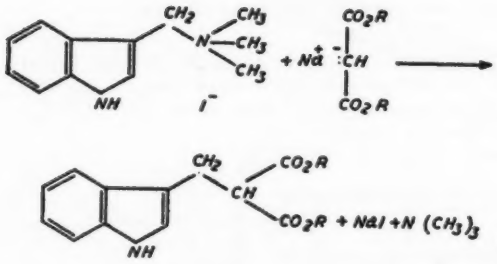
Thus the way for the tryptophane synthesis was prescribed. Starting with acetyl aminomalonate, he got skatyl-acetyl-aminomalonate, which on saponification gave tryptophane in excellent yields.

Two possible mechanisms for this reaction have been put forward by Snyder et al.[5] The first consists of an elimination of trimethylamine hydroiodide and a subsequent Michael type of addition according to that of Mannich[6] for alkylations with salts from ketonic Mannich bases (cf. equation 3).

In the second mechanism a molecule of trimethylamine is displaced from the quaternary gramine-ion by the anion of the substance being alkylated (cf. equation 4).



(3)



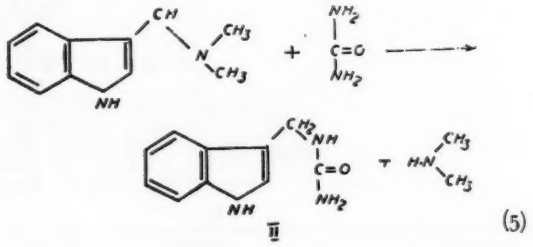
(4)

Furthermore Snyder et al. found that free gramine reacts very slowly with malonic ester but traces of alkali catalyse this process. This indicates that the reaction depends not only on the polarisation of the C-N bond in benzyl- and skatyl-amines but also on the reactivity of the hydrogen atom of the compound being alkylated. It was therefore thought worthwhile to investigate the reaction of gramine with substances containing hydrogen of various reactivity both with and without alkali as a catalyst.

The first group of substances were amides beginning with urea. By simply melting gramine in an excess of urea which served as a solvent, and keeping the temperature below 150°C. where biuret formation takes place, 46% of skatylurea (II) were formed (cf. equation 5).

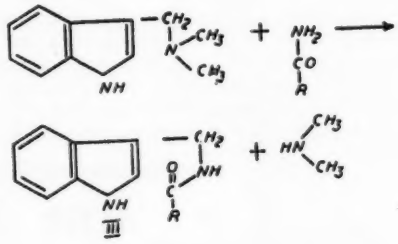
By adding a trace of alkali the yield goes up to 70%. In addition to microanalysis for all elements, the constitution of skatylurea has been confirmed by preparing the nitroso compound thus showing that skatylurea behaves like other monosubstituted ureas.

Next, acetamide, phenylacetamide, benzamide, urethane, succinamide and adipicamide were



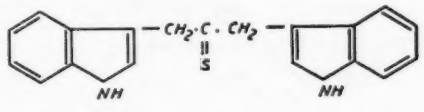
(5)

brought into reaction with gramine. Though dimethylamine was always evolved, the mixtures soon turned to dark masses out of which no crystalline material could be obtained. With traces of alkali the same reactions had already been carried out by Hellmann,[8] who found that condensation takes place with yields not more than 20%. So the idea to get compounds of structure III through this method had to be abandoned.



(6)

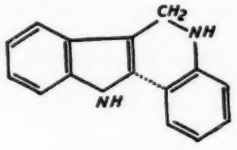
The exceptional behaviour of urea is the more remarkable as thiourea reacts in a completely different manner. Melted with gramine without alkali, sulphur containing gaseous decomposition products are evolved even at low temperature, the whole mass soon darkens and no crystalline substance could be isolated. Because urea and gramine form also skatylurea in alcoholic solution, the same conditions were used with thiourea. Soon a colourless crystalline substance came out of the clear solution which turned out to be something completely different from skatyl-thiourea.[7] Having only two nitrogens instead of three, the compound analyses for a substance to which the tentative formula of a *sym.*ββ- diindolyl-thioacetone (IV) was given.



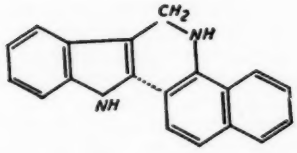
IV

7 This reference was so for not available to authors.

The peculiar behaviour of urea may be explained by the fact that urea has a remarkable basic character represented by its nitrate formation, while the amides used have more or less acidic properties, forming sodium salts. Consequently aniline, α-, and β-naphthylamine were caused to react with gramine and yielded without alkali practically quantitatively the corresponding structures V and VI, respectively. Dehydrogenation

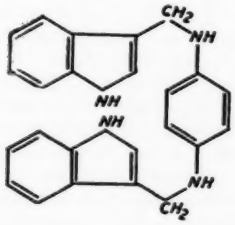


V



VI

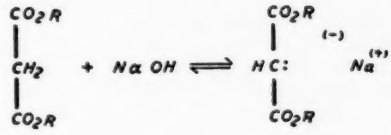
(not yet carried out), may lead to 5-carbolin systems as indicated by the dotted lines. Substituents in the *o*-, *m*-, and *p*- position to the amino-group in aniline seem to create steric hindrance in some of these compounds, e.g. *p*-phenylenediamine furnishes quantitatively the compound **VII**, while



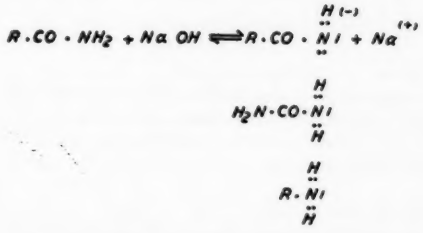
VII

o-phenylenediamine yielded only something like 4% of the *o*-isomer. The chloroanilines as well as the anisidines react in the expected manner, but *m*-nitroaniline does not react at all.

Summarising these findings, we have on the one side malonic ester and the amides as non-reacting substances; and on the other, the same substances with alkali as a catalyst and also ammonia derivatives of sufficiently high basicity, like urea, piperidine, aniline and its substitution products as reacting agents. These findings can be explained by formulating the electronic state of the reacting components. It turns out that they all have one thing in common, namely, they can be considered to be krypto-anions[8] or to become so by mesomerism under the catalytic effect of the alkali. While malonic ester and the amides are scarcely ionised, the catalytic effect of alkali favours a mesomeric state according to equation (7a) and (7b) where malonate and amide krypto-anions are formed. While any substance carrying an amino group, which is able to combine with a proton to form an ammonium salt, can be regarded to be already a krypto-anion.

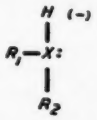


(7a)



(7b)

On this basis the reactions of benzyl- or skatyl-moiety containing quaternary ammonium salts with malonate, of free gramine with malonic ester and amides in the presence of alkali as a catalyst, and finally of free gramine with urea and amines of sufficient basicity appear to be of one and the same mechanism, namely, a replacement of a volatile krypto-anion (here dimethylamine) by another one of the general type VIII, where X can be



VIII

carbon as well as nitrogen. The extent to which this hypothesis can be generalised is subject to further investigation.

Experimental

Skatylurea.—80 g. of urea (1.3 moles) was carefully melted over a free flame and 20.2 g. of diethylskatylamine * (1/10 mole) along with

* As dimethylamine was out of stock diethylamine had to be used to prepare diethylskatylamine in Kuhn and Stein's synthesis. The m.p. 160°C. for diethylskatylamine given in the literature must be a misprint. Crystallised out of acetone, it melts at 102-

0.9 g. of powdered sodium hydroxide added. A vigorous evolution of diethylamine takes place. After about 1/2 hr. shaking and constant heating upto not more than 130°C.—because biuret-formation starts at 150°C.—all diethylskatylamine had gone in solution. The clear and slightly coloured melt was heated for an additional 2 hrs. in nitrogen atmosphere, after which time the gas evolution practically ceased. Poured in 200 ml. water the melt starts crystallising at once. Left standing overnight, filtered under suction, washed with a few ml. of ice cold water and dried on a porous plate, a first crop of 15 g. (79%) was obtained. By concentrating the mother liquor to approximately 100 ml. and adding some ml. of 2N sodium hydroxide another 2.19 g. of colourless crystals were obtained. Crude yield 17.19 g. (90.6%), m.p. 136-139°C. Recrystallised from water 13.04 g. (69%), m.p. 144-145°C. Without alkali only 46% crude yield was obtainable.

Skatylurea is easily soluble in acetone and alcohol; sparingly soluble in ethyl acetate and cold water; insoluble in benzene, carbon tetrachloride and chloroform; and recrystallisable from hot water and ethyl acetate.

Calculated for $C_{10}H_{11}O\,N_3$ (189): C, 63.5; H, 5.8; O, 8.4; N, 22.2. Found: C, 63.5; H, 5.8; O, 8.4; N, 22.3.

Nitrososkatylurea.— 1.89 g. of skatylurea (1/100 mole) and 2.5 g. of sodium nitrite were dissolved in 10 ml. of water by heating. After cooling down with ice to approximately 5°C., 1 ml. of glacial acetic acid was added. Soon the yellow needles of the nitroso compound separated. After 3/4 hrs. standing in ice the material was filtered under suction, washed with cold water and dried on a porous plate. Yield 2.16 g. (99%), decomposes at 146-150°C.

It is easily soluble in acetone; sparingly soluble in ether, petrol ether, carbon tetrachloride, chloroform and benzene; and insoluble in ethyl acetate, alcohol and dioxane. Being unstable to heating in solution, it was recrystallised by dissolving in cold acetone and precipitation with petrol ether, m.p. 150°C. dec.

Calculated for $C_{10}H_{10}O_2N_4$ (218): C, 55.0; H, 4.6; O, 14.7; N, 25.7. Found: C, 55.2; H, 4.5; O, 14.5; N, 25.5.

Gramine and Thiourea.— 1.74 g. of gramine (1/100 mole) and 0.76 g. (1/100 mole) plus 0.04 g. excess of thiourea were refluxed together with 5 ml. of alcohol in a glycerine bath heated up to 150°C. (bath temperature) for 10 hrs. On cooling, a white crystalline substance separated. Yield 1.65 g., m.p. 150-152°C. Recrystallised from alcohol, m.p. 154°C.

Calculated for $C_{19}H_{18}N\,S_2$ (306): C, 75.1; H, 5.3; N, 9.21; S, 10.5. Found: C, 74.6; H, 5.9; N, 9.3; S, 10.6.

Gramine and Amides without Alkali.— Gramine was heated in toluene on a glycerine bath at 140°C. (bath temperature) in a nitrogen atmosphere with equimolecular amounts of the following amides: acetamide, benzamide, phenylacetamide, urethane, succinamide and adipicamide. In all cases, a very slow evolution of dimethylamine took place, while the reaction mass became more and more dark and pasty. Neither short nor long heating furnished any crystalline matter except unchanged amides, while the gramine had been almost completely decomposed.

N-Skatylaniline.— 17.4 g. of gramine (1/10 mole) was heated with 40 ml. (41.3 g.) (1/10 mole plus an excess of 31.8 g.) of freshly distilled aniline in a glycerine bath under nitrogen atmosphere. Evolution of dimethylamine started at 100°C. and became vigorous at 135°C. bath temperature. Gramine slowly went into solution. The heating at 125°C. (bath temperature) was continued until the dimethylamine was no longer traceable (14 hrs.); only a slight pinkish colouration was produced. The oily mass was then steam distilled to remove the aniline. The residue was taken up in 100 ml. of benzene, dried over sodium sulphate and the benzene removed. Yield, 16.7 g. (75%); oily mass, which on standing crystallised throughout. High vaccum distillation was tried, but was not successful due to decomposition. By dissolving in seven times the volume of benzene, cooling down with ice to temperatures between 0-5°C. and adding dropwise cooled petrol ether until turbidity sets in, colourless crystals melting at 83-85°C. can be obtained. On exposure to air the substance becomes reddish. It is difficult to crystallise.

N-Skatylaniline is easily soluble in benzene, alcohol, ethyl acetate, acetone and acetic acid; and is insoluble in petrol ether and water.

Calculated for $C_{15}H_{14}N_2$ (222): C, 81.08; H, 6.30; N, 12.61. Found: C, 81.95; H, 6.36; N, 11.91.

N-Skatyl-α-naphthylamine.— 1.74 g. of gramine (1/100 mole) was heated together with 1.5 g. (1/100 mole plus an excess of 0.07 g.) of α - naphthylamine under nitrogen atmosphere

103°C. Our preparation has been confirmed by analysis.
Calculated for $C_{13}H_{18}N_2$ (202): C, 77.2; H, 8.9; N, 13.9. Found: C, 76.96; H, 9.0; N, 14.3.

in a 50 ml. round bottom flask in a glycerine bath. The mixture melted at 110-120°C. (bath temperature) and evolution of dimethylamine gas started. It was heated at this temperature until dimethylamine was no longer traceable, (30 hrs.) and kept over-night in a nitrogen atmosphere. After that time the dark brown oily mass had crystallised throughout. Yield 2.69 g.; easily soluble in benzene, ethyl alcohol, ethyl acetate, acetone and acetic acid; and insoluble in petrol ether and water. Recrystallisable out of benzene - petrol ether.

The whole mass was dissolved in 20 ml. of benzene, cooled, and ice-cold petrol ether was added dropwise until turbidity appeared (11 ml.), and was left in the refrigerator over-night. The first crop of crystals yielded 0.75 g., m.p. 148-150°C. To the mother liquor another 4 ml. of ice-cold petrol ether were added dropwise until the liquid became turbid and the mixture was kept standing over-night in the refrigerator. Another fraction of 0.89 g. could be obtained of the same m.p. (148-150°C.). Total yield 1.64 g. (60%). This pure product could be recrystallised out of benzene alone. The m.p. remained unchanged.

Calculated for $C_{19}H_{16}N_2$ (272): C, 83.82; H, 5,88; N, 10.29. Found: C, 83.72; H, 5.80; N, 10.76.

N-Skatyl-β-naphthylamine.— 1.74 g. of gramine (1/100 mole) was heated together with 1.5 g. (1/100 mole plus an excess of 0.07 g.) of β-naphtylamine in nitrogen atmosphere in a 50 ml. round bottom flask in a glycerine bath. The mixture melted at 120-130°C. (bath temperature) and the evolution of dimethylamine gas started. The gas evolution stopped practically after 27 hrs. heating, during which time small amounts of the β-naphthylamine sublimated out of the reaction mass. Solubilities and recrystallisation was the same as with the α-isomer. Total yield 1.50 g., m.p. 185-187°C. This product could also be recrystallised out of benzene alone for analysis.

Calculated for $C_{19}H_{16}N_2$ (272): C, 83.82; H, 5.88; N, 10.29. Found: C, 84.64; H, 6.28; N, 9.57.

N-Skatyl-*o*-chloroaniline.— This substance was prepared in the same way out of gramine and *o*-chloroaniline. Crude material out of benzene-petrol ether: 42%. It was recrystallised a second time out of benzene- petrol ether and gave a m.p. of 70-72° C.

Calculated for $C_{18}H_{13}N_2Cl$ (256.5): C, 70.18; H, 5.06; N, 10.91; Cl, 13.84. Found: C, 70.43 H, 5.51; N, 10.54; Cl, 13.34.

N-Skatyl-*p*-chloroaniline.— Gramine and *p*-chloroaniline yielded 36% of the condensation product from benzene-petrol ether, m.p. 98-100°C.

Calculated for $C_{18}H_{13}N_2Cl$ (256.5): C, 70.18; H, 5.06; N, 10.91; Cl, 13.84. Found: C, 71.39; H, 5.28; N, 10.49; Cl, 12.88.

N-Skatyl-*m*-chloroaniline.— The reaction between gramine and *m*-chloroaniline was carried out in the same manner as described before. By recrystallisation from benzene and petrol ether only an oily substance could be obtained in spite of repeating the procedure seven times with long standing in the refrigerator. Due to the sensitivity against acids—all these skatyl derivatives become red at once—salt formation could not be used for further purification.

N-Skatyl-*m*-nitroaniline.—From the reaction between gramine and *m*-nitroaniline too, dimethylamine was developed, but only dark reddish decomposition products could be observed to have been formed.

N,N′-Diskatyl-*p*-phenylenediamine.—3.44 g. of gramine (1/50 mole) and 1.08 g. of *p*-phenylenediamine (1/100 mole) had been well mixed in a mortar and heated in nitrogen atmosphere in a glycerine bath. The mixture melted at 130°C. and the development of dimethylamine followed almost immediately. After 5 hrs. heating at 135°C. (bath temperature) 80% of the theoretical amount of dimethylamine had been evolved, as determined by repeated weighing of the reaction flask. The gas evolution was then very slow and the experiment was stopped. The crude mass melted at 170-175°C. By treating with a few drops of benzene, the melt recrystallises and 2.7 g. (73.7%), m.p. 174-176°C., could be obtained. After recrystallisation twice from ethyl acetate, the melting point was constant at 183-185°C. The substance is easily soluble in acetic anhydride (acetylation), sparingly soluble in petrol ether, ethyl acetate, acetone, ethyl alcohol; insoluble in benzene; and recrystallisable from ethyl acetate.

Calculated for $C_{24}H_{22}N_4$ (366): C, 78.68; H, 6.01; N, 15.3. Found: C, 78.6; H, 5.99; N, 15.47.

N,N′-Diacetyl-N,N′-diskatyl-*p*-phenylene-diamine.— 1.5 g. of N,N′-diskatyl-*p*-phenylene-diamine was spread in a thin layer in a petri dish and just enough acetic anhydride was added to moisten the substance. With development of

heat the substance goes in solution, and almost immediately the acetylated compound separated in well shaped prisms so that the whole mass became solid. After some standing and cooling, it was filtered under suction and washed with a few ml. of cooled anhydride, yield 1.5 g. (76.6%).

The acetyl compound is sparingly soluble in glacial acetic acid, dioxane, ethyl alcohol and benzene; and recrystallisable by dissolving in glacial acetic acid with heating and after some concentration precipitating with ethyl acetate, m.p. 250-252°C.

Calculated for $C_{28}H_{26}O_2N_4$ (450): C, 74.66; H, 5.77; N, 12.44; O, 7.11. Found: C, 73.94; H, 5.76; N, 11.65; O, 8.46.

N,N′ -Diskatyl - *o* - phenylenediamine.— This reaction was carried out in the same way as with the *p*-phenylenediamine, but very little crystalline substance could be isolated from the oily reaction product. This substance (in about 5% yield) melted at 185-195°C., and behaved in the same manner against acetic anhydride as the para-isomer furnishing 4% of theoretical yield of acetylation product, m.p. 221-223°C. from ethyl acetate.

Calculated for diacetyl product $C_{28}H_{26}O_4N_5$ (450): C, 74.66; H, 5.77; N, 12.44; O, 7.11. Calculated for tetraacetyl product $C_{32}H_{30}O_4N_4$ (534): C, 71.9; H, 5.4; N, 10.4; O, 11.9. Found: C, 71.34; H, 6.15; N, 12.63; O, 9.71.

N-Skatyl -*p*- anisidine.— 3.48 g. of gramine (1/50 mole) and 2.4 g. of *p*-anisidine (1/50 mole) were well mixed in a mortar and heated on a water bath. Dimethylamine developed as soon as the mixture melted and continued to come out during 17 hrs. After that the mass crystallised on cooling. By making a paste with ethyl alcohol, the oily impurities were removed. A colourless material 2.5 g. (50%) was obtained, m.p. 94-99°C.

It is easily soluble in acetone; sparingly in ether, benzene, ethyl acetate, ethyl alcohol and chloroform; insoluble in petrol ether; and recrystallisable from ethyl alcohol, m.p. 188-190°C.

Microanalyses were carried out by A. Bernhadrt, Max Plank Institut, für Kohleforschung, 22 a Mulheim (Ruhr), W. Germany. Substances were dried over phosphorus pentoxide at 50°C. in a high vacuum before analysis.

References

1. H. v. Euler and H. Hellstroem, Zeitschr. physiolog. Chem., **208**, 43 (1932).
2. A. P. Orekhov, S.S. Norkina and T. Maximova, Ber., **68**, 436 (1935); H.v. Euler, E. Erdtman and H. Hellstroem, Ber., **69**, 743 (1936); A.P. Orekhov and S.S. Norkina, J. Gen. Chem. USSR, **7**, 673 (1937).
3. Kuhn and Stein, Ber., **70**, 567 (1937).
4. H.R. Snyder et al., J. Am. Chem. Soc., **66**, 350,500 (1944).
5. H.R. Snyder and E. Eliel, J. Am. Chem. Soc., **70**, 1703 (1948).
6. C. Mannich, G. Ball, and Reistert, Arch. Pharm., **264**, 65, 164, (1926).
7. H. Hellmann, Angew. Chem., **65**,473 (1953).
8. H. Meerwein, Liebigs Ann. Chem., **455**, 227 (1927).

PREVENTION OF MALARIA TRANSMISSION WITH SINGLE DOSE OF PYRIMETHAMINE (DARAPRIM)

COLONEL M.K. AFRIDI AND ABDUL RAHIM

North Regional Laboratories, Pakistan Council of Scientific and Industrial Research, Peshawar

Introduction

The success of residual insecticides in the prevention of malaria has been convincingly demonstrated in many parts of the world. Under certain conditions, however, this method has not given the desired results, either because the outdoor resting habits of the anopheline carrier enabled it to avoid contact with the lethal film, *e.g.*, *A. sergenti* in Jordan,[1] or because facilities for undertaking organised and widespread sprayings were too meagre to permit an adequate coverage of an intensely malarious region, *e.g.*, Equatorial Africa. Even in countries where malaria has been almost completely eradicated, the disease tends, for various reasons, to persist in small foci despite intensive sprayings. Lastly, because of the occurrence of resistance in anophelines to insecticides it is now considered necessary to eradicate malaria completely and speedily from a community while insecticides are still effective against the local carriers.

To meet these and allied situations, it is clearly desirable to develop an anti-malaria measure which would supplement residual insecticides or if necessary replace them. A field trial was accordingly planned to test whether the prolonged sporontocidal property of pyrimethamine could be utilized to serve this purpose.

Basic Consideration

To be successful, chemoprophylaxis requires a highly efficient organization for the repeated administration of drug. In this respect the situation has eased to some extent latterly as a number of the newer anti-malarials need be given only once a week or once a fortnight. Under rural conditions, however, the regularity of drug administration cannot be assured even at such intervals and, the scope of collective drug prophylaxis has, for this reason, remained considerably restricted. The situation, however, appeared more hopeful when one of us (M.K.A.) received a communication from Dr. G.R. Coatney[2] to the effect that "on a calculated basis following a single 25 mg. dose of pyrimethamine there would still be a suppressive amount (*i.e.*, 0.8 mg. of the drug) in the body after 52 days." The inference drawn from this observation was that if the amount remaining in the body proved effective against sporogenic cycle then a single dose of pyrimethamine would function as a "residual sporontocide" for at least 52 days. This time period, if confirmed in the field, would not only be much longer than that anticipated by Roberts[3] but would be sufficient to make the successful application of chemoprophylaxis in rural areas a feasible proposition.

Description of the Trial

Since the main object of the trial was to trace the effect of pyrimethamine on malaria transmission, it was felt that this purpose would be best served by making repeated observations on a small stable community. The trial, which lasted from July 1st to mid-October, was therefore, organized on a population of 1,289 in a group of four villages in the valley between Haripur and Havelian in the Hazara district of West Pakistan. Unlike the compact villages usual in this part of the country, the test villages consisted of a series of hamlets strung out in a row on the bank of a small river. A single dose of Daraprim (pyrimethamine) was given to the inhabitants of three of the villages namely Chamba, Dhaia and Dobandi, located in that order from north-east to south-west over a distance of about 3 miles, while the fourth village, called Nika Pa, which was the southern-most in the experimental area, received no treatment and was kept as control. Not only was spraying disallowed in these villages during the test season, but one of the factor that influenced their selection was that they had not been sprayed in previous years. The latter consideration greatly restricted our field of choice as spraying operations had been fairly widespread in this area. Another factor kept in mind was to select a more or less isolated block of villages so that the results would not be vitiated by the incursion of infected mosquitoes from the adjoining untreated areas. In this respect Chamba and Dhaia were ideally placed but Dobandi was not as the distance between the latter and the untreated village Nika Pa was less than ½ mile.

The drug was given in accordance with the dosage scale of 25 mg. to adults, 12.5 mg. to children from 7 to 12 years of age and 6.25 mg. to children aged 6 years and below. Particulars of every individual in the test and control villages were noted in separate registers and the dosage given to each person was recorded at the time

TABLE I.—POPULATION AND TREATMENT FIGURES.

Name of Village	Population	Number received treatment	Number who avoided treatment	Percent missed treatment
TEST				
Chamba	601	592	9	1.5
Dhaia	274	262	12	4.4
Dobandi	186	160	26	14.0
CONTROL				
Nika Pa	228	Nil.	—	—

the drug was administered. This procedure ensured not only a reliable record of pyrimethamine administration, but also an up-to-date census of the population under test.

Every effort was made to administer the drug to the maximum number of persons. In this respect we were fairly successful, as will be seen from Table I which shows that the percentage of persons who avoided treatment was reasonably low except in Dobandi.

Assessment

To assess the effects of pyrimethamine, malariometric data and figures for malaria incidence were collected simultaneously from all the villages under observation.

Spleen rates were taken at the beginning and again at the end of the malaria season in mid-July and mid-October, respectively, but examinations for parasites were conducted every month from July to October. Special efforts were made to examine the maximum number of infants, as it was felt that the occurrence or otherwise of malaria transmission could be best deduced from infection in this group, there being no facilities for mosquito dissection.

As regards malaria cases, two methods were used for obtaining the figures. First, a treatment centre was established in a centrally placed village which was accessible equally to all the inhabitants of the experimental area. Every patient reporting at this centre with fever had a blood slide examined for malaria parasites and was given a single dose of amodiaquine (Camoquine) in the scale, 600 mg. of active base to adults, 400 mg. to children from 5 to 15 years, 200 mg. to children under five years. Camoquine was selected for clinical treatment, because it was considered least likely to affect the sporontocidal action of pyrimethamine. The treatment centre proved very popular attracting nearly all the malaria cases that occurred in the neighbourhood.

The second set of figures was collected during special rounds when all the villages were searched on one specified day for malaria cases by house-to-house enquiry. Unfortunately, the idea of undertaking such rounds, which came to be designated "Malaria Census," struck us late in the season, the first one being on the 5th of September, while the three subsequent rounds were carried out regularly every two weeks.

Malariometric Data

In Table II are given the spleen rates of children from three to nine years of age and in Table III parasite rates in children and youths of varying age groups.

From a study of the July spleen rates in Table II, it will be seen that although the rates in different villages varied, all of them belonged to the meso-endemic class. The spleen rates increased progressively in the villages from north to south, indicating that the northern group, comprising Chamba and Dhaia, was relatively less malarious than the southern group of Dobandi and Nika Pa. Particular importance was, however, attached to the latter two villages inasmuch as they were both equally malarious, and any variation in the rates during or after the malaria season could

TABLE II.—SPLEEN RATES IN CHILDREN FROM 3 TO 9 YEARS OF AGE.

Name of Village	JULY					OCTOBER				
	No. exam.	No. positive	Spleen rates	A.E.S.	S.I.	No. exam.	No. positive	Spleen rates	A.E.S.	S.I.
TEST—										
Chamba ..	158	24	15.18	1.5	22.77	132	4	3.03	1.0	3.03
Dhaia ..	79	18	22.78	1.8	41.00	81	8	9.87	1.0	9.87
Dobandi ..	56	24	42.75	1.5	64.13	52	6	11.53	1.7	19.60
CONTROL—										
Nika Pa ..	69	28	40.58	1.4	56.84	65	25	38.46	1.2	46.15

A. E. S. — Average enlarged spleen. S. I. — Splenometric Index.

TABLE III.— PARASITE RATES.

MONTH AND AGE GROUPS	CHAMBA			DHAIA			DOBANDI			NIKA PA (Control)		
	No. examined	No. positive	Parasite rate	No. examined	No. positive	Parasite rate	No. examined	No. positive	Parasite rate	No. examined	No. positive	Parasite rate
JULY												
0 to 1 year ..	17	0	0	6	0	0	4	0	0	3	1	—
1 to 9 years ..	195	10	5.13	94	4	4.26	44	3	6.82	65	7	10.77
10 to 16 years ..	53	0	0	18	0	0	8	0	0	15	2	13.33
AUGUST												
0 to 1 year ..	5	0	0	3	0	0	3	0	0	3	1	—
1 to 9 years ..	138	5	3.62	60	4	6.67	49	3	6.12	59	10	16.95
10 to 16 years ..	129	0	0	12	0	0	6	0	0	11	1	9.09
SEPTEMBER												
0 to 1 year ..	9	0	0	6	0	0	7	1	14.29	2	2	—
1 to 9 years ..	161	4	2.48	80	4	5.00	58	7	12.07	47	11	23.40
10 to 16 years ..	27	0	0	13	0	0	9	0	0	7	2	28.29
OCTOBER												
0 to 1 year ..	10	0	0	6	0	0	7	0	0	3	0	0
1 to 9 years ..	160	1	0.63	97	0	0	63	2	3.17	68	7	10.29
10 to 16 years ..	52	0	0	23	0	0	20	0	0	13	0	0

only be occasioned by the effects of pyrimethamine.

In the control village Nika Pa, the spleen rates as well as splenometric indices displayed a minor fall in October instead of the expected rise, due probably to the prompt treatment of malaria cases. In the test villages, on the other hand, the fall in October rates was considerably steeper which could have been brought about only by the interruption of transmission. This is clear from a study of the ratio of July to October splenometric indices which in the control village was 1.2:1 against 7.5:1, 4.2:1 and 3.3:1 in the test villages Chamba, Dhaia and Dobandi, respectively.

Table III shows that malaria infection (*P. vivax*) occurred in one infant in the control village in July, actually on the second day of that month. Malaria transmission was, therefore, already taking place in the experimental area when pyrimethamine was being distributed. In August the same infant was again found to harbour *P. vivax* but in September two other infants in this village were infected, one with *P. vivax* and the second with *P. falciparum*. Thus, out of a total of four infants in the control village, malaria infection occurred in three while the fourth might or might not have been infected as he could not be examined in September and October.

In contrast to this, there was no infection in infants in Chamba and Dhaia, indicating the absence of transmission in these two test villages. In Dobandi, on the other hand, *vivax* infection was found in one infant on 7th September, *i.e.*, in the tenth week after the distribution of the drug. Allowing for the two incubation periods, this infection may be traced to potent gametocytes that were circulating in the blood round about the second week of August. The sporontocidal effects of a single dose of pyrimethamine can thus be reckoned to have lasted for approximately 42 days which is ten days short of the time-period given by Coatney.[2]

The parasite rates amongst older children in the different villages conf rmed to the findings in infants. Thus, in the control village, the rates in children aged 1 to 9 years displayed a definite rise in August which ended in a peak in mid-September. Species distribution of *P. vivax* and *P. falciparum* shown in Table IV was also of the order usually encountered in a normal seasonal malaria outbreak.

In the test villages, Chamba and Dhaia, the rates in the same age groups remained stationary in August while in September instead of a rise they displayed a fall. This finding together with the fact that no *P. falciparum* infection was found in either of the villages was an added evidence pointing to complete interruption of transmission. In Dobandi, on the other hand, the August rates were low but in September not only did the rates rise markedly but also there was one *P. falciparum* infection. Unlike control village, Nika Pa, however, the steep September rise in Dobandi was followed by an equally precipitous fall in the rates in October indicating that transmission in the latter village supervened too late in the season to bring about the usual cumulation of malaria infection and the rise in *P. falciparum*.

Turning to the parasite rates in the age group 10-16 years it was not always possible to sample an adequate number of persons. However from a study of the available figures in Table III it will be seen that only the control village showed infection in this group.

Incidence of Malaria

Figures for malaria cases from the two sources, namely, the treatment centre and the malaria census, are shown in Table V. These figures portray almost the same picture as that presented by the parasite rates. For here also, the incidence of the disease was low in Chamba and Dhaia, higher in Dobandi but much higher in the control village. Species distribution also conformed to the pattern seen in the parasitic survey, all cases in Chamba and Dhaia being benign tertian while in Dobandi two out of 15 cases were malignant tertian. In the control village, on the other hand, *P. falciparum* prevailed in a much higher proportion as will be seen from the figures presented in Table VI.

Discussion

Judging from the results of observations on the control village, malaria may be said to have prevailed in the experimental area in a moderately severe form during the season under review. It is, therefore, particularly significant that, in such a year, a single dose of pyrimethamine should have succeeded in eliminating transmission in two out of thr.e test villages throughout a season of 3½ months. In the third test village, Dobandi, a break-through occurred about 42 days after the administration of pyrimethamine but conditions there were peculiar. In the first place, no less than 14 per cent of its inhabitants had missed the drug, vide Table I. Secondly, owing to its location, the village was exposed to the invasion of infected mosquitoes from the adjoining untreated sector to a greater extent than the other two test

TABLE IV.—SPECIES DISTRIBUTION OF *P. vivax* AND *P. falciparum* IN CHIDREN AGED 1 TO 9 YEARS IN THE CONTROL VILLAGE.

Niki Pa	*P. vivax*	*P. falciparum*	Mixed *vivax* and *falciparum*	Total
July	9	1	..	10
August	10	1	1	12
September	9	5	1	15
October	3	4	..	7
	31	11	2	44

TABLE V.—INCIDENCE OF MALARIA.

Name of village	Population	Cases treated in Treatment Centre					Cases found during Malaria Census					Total from cases both sources	Incidence ratio per 1000 population
		July	Aug.	Sept.	Oct.	Total	5th Sept.	19th Sept.	3rd Oct.	16th Oct.	Total		
TEST—													
Chamba	601	0	1	1	—	2	1	1	1	0	3	5	8.32
Dhaia	274	0	1	0	0	1	1	3	0	0	4	5	18.25
Dobandi	186	0	1	9	2	12	2	1	0	0	3	15	80.65
CONTROL—													
Nika Pa	228	1	8	16	6	31	5	3	3	1	12	43	188.60

TABLE VI.—SPECIES DISTRIBUTION IN MALARIA CASES IN THE CONTROL VILLAGE NIKA PA.

Species	Treatment Centre Figures					Malaria Census Figures				
	July	August	Sept.	Oct.	Total	5 Sept.	19 Sept.	3 Oct.	16 Oct.	Total
P. vivax	—	6	7	2	15	4	2	1	—	7
P. falciparum	—	1	7	4	12	1	1	2	1	5
P. malariae	1	—	—	—	1	—	—	—	—	—
Mixed *P. vivax* and *P. falciparum* ..	—	1	2	—	3	—	—	—	—	—
	1	8	16	6	31	5	3	3	1	12

villages. Thirdly, it had the highest spleen rates indicating that pyrimethamine was probably exposed to the severest infection pressure there.

From the available evidence it is impossible to deduce which of these factors, singly or jointly, caused the break-through of transmission in Dobandi. The question is, however, of considerable practical importance and deserves further investigation as connected with it is the problem whether higher dosage or more frequent administration of pyrimethamine than that used in this trial will be necessary to prevent a break-through of transmission in villages with high spleen rates. Satisfactory results have been recorded from a single dose of pyrimethamine (Daraprim) in a hyperendemic area in Kenya[4] but in that trial the sporontocidal action of the drug was not especially investigated.

Our trial has the merit of demonstrating the value of sporonotocide as a preventive of malaria transmission but only in an area of moderate endemicity with a short malaria season. We recognize that pyrimethamine has the risk of uncovering resistant strains of parasites but the potential advantages of the method are so patent that it deserves an extensive trial despite the possibility of this complication. To reduce the chance of resistance, this drug may perhaps, be reserved exclusively for malaria prevention as pointed out by Covell et al., [5] although it would be difficult to say how effective this precaution would be considering that a single dose of the drug has been reported to have caused the appearance of a resistant strain in Kenya.[4]

Summary

1. A field trial to test the utility of a single dose of pyrimethamine as preventive of malaria transmission is described. The dosage scale given was 25 mg. to adults, 12.5 mg. to children from 7 to 12 years of age, and 6.25 mg. to children aged 6 years and below.

2. The trial covered a small community kept under continuous observation over a malaria season. No evidence was forthcoming of malaria transmission in two out of the three test villages for 3½ months but transmission broke through in the third village.

3. Under the conditions of the trial, the sporontocidal action of the single dose of pyrimethamine was found to last a minimum of 42 days even in the most adverse local circumstances.

4. It is concluded that this method of control deserves trial in other areas as its potential advantages outweigh its possible drawbacks, namely, the practical difficulties in organizing mass chemoprophylaxis and the risk of bringing resistant strains to surface.

Acknowledgement

We wish to thank Dr. Salimuzzaman Siddiqui, Director, Pakistan Council of Scientific and Industrial Research, for his stimulating criticism and for permission to publish this paper.

References

1. M.A. Farid, Bull. World Health Organization, **11** (4-5), 675-783 (1954).
2. G.R. Coatney, National Institute of Health, Bathesda, U.S.A., personal communication.
3. J.M.D. Roberts, J. Trop. Med. Hyg., **59**, 201-8 (1956).
4. S. Avery Jones, E. African Med. J., **31**, 47-49 (1954).
5. G. Covell, G.R. Coatney, J.W. Field and J. Singh, *Chemotherapy of Malaria*, World Health Organization, Monograph series No. 27 (Geneva, 1955).

EVALUATION OF FUNGICIDAL PROPERTIES OF LIGNIN ACETIC ACID

Part II.—Studies on Treated Jute Seeds in Infested Soil

M.I. Ali, Q.A. Ahmed and M.H. Khundkar

Department of Chemistry, University of Dacca, Dacca, East Pakistan

In view of the promising results obtained in the previous work,[1] elaborate studies were undertaken on jute seeds differently treated with lignin acetic acid for controlling the diseases of jute plants caused by various fungus organisms. Preliminary experiments were carried out with a sample of C.G. variety of *Corchorus olitorius* that showed 14.0% contamination with *Macrophomina*. The seeds were surface sterilized with 0.5% solution of lignin acetic acid as seed protectant and allowed to germinate on wet filter paper in a sterilized petri-plate. The result was satisfactory in so far as these seeds have shown no evidence of toxic effect upon the germination and the extent of the disease was brought down from 14.0 to 2.0%. This inspired further study of the problem, particularly in disease infested soil.

Experimental

The detailed experiments relating to the control of fungal diseases of jute plants were done with only two organisms, *viz.*, *Rhizoctonia sp.* and *Macrophomina phaseoli*, which are reported[2] to cause heavy damage to the seedlings.

I. Preparation of treated seed samples : A sample of C.G. variety of *Corchorus olitorius* was treated with lignin acetic acid in three different ways as described below. Fifteen samples of treated seeds (five for each treatment) and one of untreated seeds were stored for about 6 months for investigation.

Treatment 1 : The alcoholic solution of lignin acetic acid was diluted with distilled water to a large volume to make 0.5% and 1.0% solutions separately. The seeds were surface sterilized with this solution, dried and stored carefully.

Treatment 2 : A 5.0% water suspension of lignin acetic acid was prepared and the seeds were treated with it, dried and stored.

Treatment 3 : A dry powder of lignin acetic acid (6.6 % by wt.) was added to the seeds and mixed up intimately by mechanical means, after which the seeds were stored.

II. Preparation of infested soil : The sterilized soil was kept open for about 7 days to vent the unwanted gases. The soil was then made properly wet and the requisite quantity was taken in several earthen pots. Previously grown fungus organisms* were mixed with the required quantity of sterilized soil by a mechanical process. A ¾ inch layer of this inoculated soil was spread over the sterilized soil previously filled in the pots. The pots—now containing two layers of soil, the upper layer profusely contaminated with the organism and the lower layer without pathogen—were kept aside for 7 days to obtain a uniform distribution of fungus throughout each pot, after which they were ready for sowing.

III. Sowing and collection of data: It was felt necessary to keep 5 pots for each sample of treated and untreated seeds, 10 seeds being sown in each pot. The pots were kept at room temperature (about 27 °C.), the requisite quantity of water being supplied regularly. The number of seeds germinated, as well as those dying before and after germination were noted every day for the first 5 days, and thereafter every fifth day. The final results after 30 days are presented in Tables 1, 2 and 3.

From the tables, it seemed that about 10% death always occurred with both treated and untreated seeds in sterilized soil without any pathogen. Sometimes the plants appeared to be healthier with treated seeds. In the case of untreated seeds in *Rhizoctonia* infested soil, the death record was 64% whereas with the treated seeds it has been brought down to 8-12% in different samples. In experiments with *Macrophomina*, on the other hand untreated seeds showed 42% deaths, which were lowered to 26% in the treated seeds. As far as could be observed, lignin acetic acid showed no toxic effect upon the germination.

Discussion

On the basis of statistical probability the results can be tested for significance as follows:—

$$\frac{P_1 - P_2}{\sqrt{\sigma_1^2 + \sigma_2^2}} \geqslant 2 \text{ for definite significance,}$$

* Test orgunism grown for 7 days in sterilized medium prepared as follows : 100 g. rice-husk ; 2.5 % glucose ; 4.7 % ammonium sulphate; and 30 cc. water.

Table 1.—Germination of Variously Treated Jute Seeds in Different Potting Soils.

Nature of inoculum and treatment	1			2			3			4			5			Remarks
	G	PrD	PoD	G	PrD	PoD	G	PrD	PoD	G	PrD	PoD	G	PrD	PoD	
Control sterilized oil, without inoculum and untreated seeds used	10	—	1	10	—	2	10	—	1	10	—	1	10	—	—	G =50 *i.e.* 100% PrD= 0 0 PoD= 5 *i.e.* 10%
Sterilized soil without inoculum and treated seeds (1.0% alcoholic solution)	10	—	1	10	—	1	10	—	1	10	—	1	9	1	1	G =49 *i.e.* 98% PrD = 1 *i.e.* 2% PoD= 5 *i.e.* 10%
Sterilized soil without pathogen and treated seeds (5.0% water solution)	10	—	1	9	1	—	9	1	1	10	—	1	10	—	1	G =48 *i.e.* 96% PrD = 2 *i.e.* 2% PoD= 4 *i.e.* 8%
Sterilized soil without pathogen and treated seeds (6.6% dry powder)	10	—	1	10	—	1	9	1	—	10	—	—	10	—	1	G =49 *i.e.* 98% PrD = 1 *i.e.* 2% PoD= 3 *i.e.* 6%

Table 2—Germination of Variously Treated Jute Seeds in Soil Infested with *Rhizoctonia sp.*

Nature of inoculum and treatment	1			2			3			4			5			Remarks
	G	PrD	PoD	G	Prd	PoD	G	PrD	PoD	G	PrD	PoD	G	PrD	PoD	
Steelized soil infested with *Rhizoctonia sp.* and untreated seeds.	9	1	7	9	1	6	10	—	5	10	—	5	10	—	7	G = 48 *i.e.* 98% PrD = 2 *i.e.* 4% PoD = 30 *i.e.* 60%
Sterilized soil infested with *Rhizoctonia sp.* and treated seeds (1.0% alcoholic solution)	9	1	—	9	1	—	10	—	1	9	1	1	9	1	—	G = 46 *i.e.* 92% PrD = 4 *i.e.* 8% PoD = 2 *i.e.* 4%
Sterilized soil infested with *Rhizoctonia sp.* and treated seeds (5.0% water suspension)	10	—	—	9	1	—	10	—	1	10	—	—	9	1	1	G = 48 *i.e.* 96% PrD = 2 *i.e.* 4% PoD = 2 *i.e.* 4%
Sterilized soil infested with *Rhizoctonia sp.* and treated seeds (6.6% dry powder)	9	1	—	10	—	—	9	1	—	9	1	—	10	—	1	G = 47 *i.e.* 94% PrD = 3 *i.e.* 6% PoD = 1 *i.e.* 2%

Table 3.—Germination of Variously Treated Jute Seeds in Soil Infested with *Macrophomina.*

Nature of inoculum and treatment	1			2			3			4			5			Remarks
	G	PrD	PoD	G	PrD	PoD	G	PrD	PoD	G	PrD	PoD	G	PrD	PoD	
Sterilized soil infested with *Macrophomina* and untreated seeds	8	2	8	9	1	—	10	—	2	10	—	2	9	1	5	G = 46 *i.e.* 92% PrD = 4 *i.e.* 8% PoD = 17 *i.e.* 34%
Sterilized soil infested with *Macrophomina* and treated seeds (1.0% alcoholic solution)	9	1	2	9	1	1	9	1	2	8	2	2	10	—	3	G = 45 *i.e.* 90% PrD = 5 *i.e.* 10% PoD = 10 *i.e.* 20%
Sterilized soil infested with *Macrophomina* and treated seeds (5.0% water suspension)	9	1	2	9	1	1	10	—	3	8	2	1	10	—	2	G = 46 *i.e.* 92% PrD = 4 *i.e.* 8% PoD = 9 *i.e.* 18%
Sterilized soil infested with *Macrophomina* and treated seeds (6.6% dry powder)	8	2	2	9	1	3	8	2	1	9	1	2	10	—	3	G = 44 *i.e.* 88% PrD = 6 *i.e.* 12% PoD = 11 *i.e.* 22%

G—Number of seeds germinated ; PrD—Pre-emergent death; PoD—Post-emergent death.

where P_1 and P_2 are the proportions of death in the control and the other experiment in question, and σ_1 and σ_2 are the variances of P_1 and P_2 respectively. The value for the above experiments are given in Table 4.

TABLE 4.—SIGNIFICANCE TEST FOR EFFECTIVENESS OF TREATMENT AGAINST RHIZOCTONIA AND MAGROPHOMINA

(Values of $(P_1-P_2)/\sqrt{\sigma_1^2+\sigma_2^2}$ are given)

Type of treatment with lignin acetic acid	*Rhizoctonia*	*Marcophomina*
1 % alcoholic solution	6.36	1.26
5 % water suspension	7.23	1.71
6.6 % dust suspension	7.23	1.02

In view of the above results it appears that all the results obtained against *Rhizoctonia* are significant (*i.e.*, greater than 2), but in the case of *Macrophomina* the results are insignificant or at best barely significant.

Thus we may conclude from our experiments carried out on *Rhizoctonia sp.* and *Macrophomina* infestation in soil that the treatment of seeds with (i) 1.0% alcoholic solution, (ii) 5.0% water suspension or (iii) 6.6% dust of lignin acetic acid is entirely effective in controlling the diseases caused by *Rhizoctonia sp.*, the mortality being brought down from 64% to 8-12%. The results against *Macrophomina*, although promising cannot be constituted as statistically significant.

Acknowledgement

The investigation was carried out under a research scheme on "Studies on Lignin," financed by the Pakistan Council of Scientific and Industrial Research.

A part of the work was done in the Mycology Section of the Department of Botany and our thanks are to due to Prof. M. Ahmad for the courtesy.

Our thanks are also due to Messrs. Karnaphuli Paper Mills Ltd., Chandraghona, for the supply of black liquor.

References

1. M.I. Ali, Q.A. Ahmed and M.H. Khundkar, Pakistan J. Sci. Ind. Research, **1**, 79 (1958).
2. K.M. Badruddoza, Krishi-Katha, **14**, No. 2, 120 (1954).

STUDIES IN THE RELATIONSHIP BETWEEN VISCOSITY AND MOLECULAR STRUCTURE

Part I.—Temperature Dependence of the Viscosity of certain Typical Mineral Oils.

MAZHAR MAHMOOD QURASHI

Central Laboratories, Pakistan Council of Scientific and Industrial Research, Karachi

1. Introduction

For some time past, the behaviour of mixtures of mineral oils with various hydroxylic liquids has been the subject of investigation in this laboratory.[1] On examining the results obtained it was thought worth-while to undertake a detailed study of the temperature-dependence of the viscosities of mineral oils as well as hydroxylic compounds. Because these liquids have high viscosities and are usually considered to be more or less anomalous, it was expected that their study would give valuable information about the correlation between viscosity and molecular structure. In general terms, we may picture such a liquid as composed of molecular aggregates of varying size and orientation, each with exposed hydroxyl (or other polar) groups capable of forming some sort of "transitory bond" with any neighbouring aggregates. The viscosity would then be dependent both on the size of the aggregates and on the strengths of these "bonds" between them. One or both of these quantities may vary with temperature, and therefore a detailed study of the temperature variation of the viscosity should give quantitative information about these characteristics of the aggregates.

There is also a more practical aspect of the problem, namely that of predicting the viscosity of a given oil or other liquid at elevated temperatures or at very low temperatures. The establishment of a simple generalized formula for this could be of considerable utility in the field of lubricants, anti-freeze compounds, etc.

2. Preliminary Experiments

The earliest results were obtained with a commercial straight mineral oil cut, B.O.C. "450," which has a viscosity of 400 centistokes at a temperature of 40°C., and corresponds to a fairly heavy lubricating oil. The kinematic viscosities were measured at different temperatures with a U-tube viscometer of the British Standard pattern mounted vertically in an electrically controlled thermostatic bath, the measurements being carried out in accordance with the Standard Specifications. The final accuracy of the determinations at each temperature was of the order of 0.2%. The results are given in Table 1(a) at intervals of 5°C. to 10°C., over the temperature range 20°—85°C. An attempt was first made to obtain a linear plot against temperature by means of a power-law, as has been done for several non-associated liquids cf. for example Mitra.[2] It was found that $1/v^{\frac{1}{4}}$ gave good results, as shown by the straight line through the observed data in Fig. 1(a). The fit is remarkably good, except for the two initial points, both of which lie above the line, suggesting a systematic divergence at low temperatures.

In order to follow up this phenomenon by working at lower temperatures, a small 2-litre thermostat was built incorporating a copper cooling coil, through which ice-cold water could be circulated at a regulated speed. In this way, it was possible to work at temperatures down to 4°C., the degree of constancy attained during each reading was however poorer than with the electrically controlled apparatus, being now only of the order of $\pm$ 0.2°C. The additional observations obtained in this way are shown in Table 1(b), and are plotted in Fig. 1(b) as hollow circles. It is seen that the complete data can be fitted almost exactly by means of two straight lines intersecting at a temperature of about 30°C., the slopes of the two lines are 0.112/°C. and 0.135/°C., respectively. The experimental point at 30°C. lies a little above both lines, but can be made to fit over the temperature range 25°C. to 35°C., if a gradual transition from one line to the other is assumed, cf. the broken line in Fig. 1(b). At all events, these graphs show evidence of some sort of transition in the viscous behaviour of the oil at a temperature of about 28°C. and a viscosity of nearly 1,000 centistokes.

The investigation was next carried over to the light lubricating oils by working with another straight mineral oil cut, B.O.C. "60," which has a viscosity of 38 centistokes at 40°C. The experimental values are given in Table 1(c), while the upper graph of Fig. 1(b) shows that these values can again be represented quite well by two straight lines with a fairly sharp change-over in the region of 55°C. The slopes of the two lines are 0.157/°C. and 0.133/°C. in this case, the direction of the knee being opposite to that in the first case. It

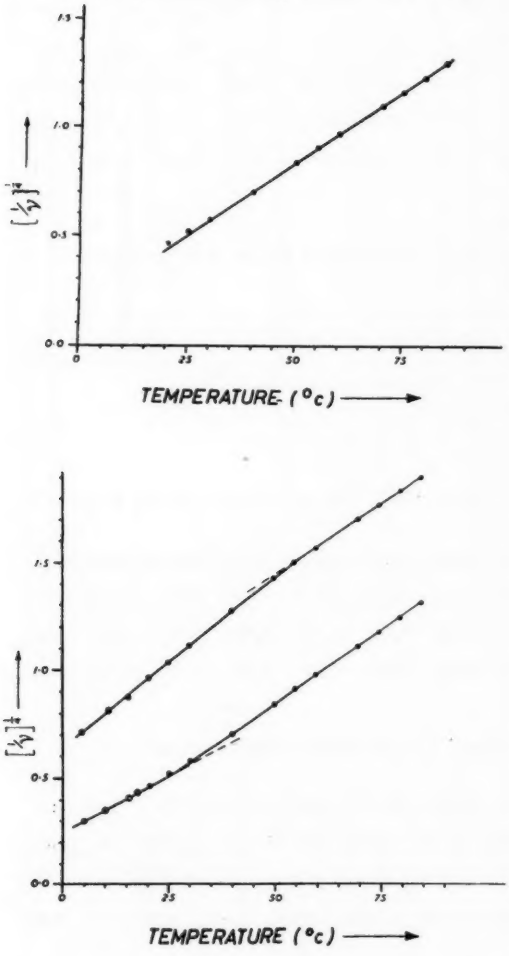


Fig. 1.—Preliminary graphs of $(1/\nu)^{\frac{1}{2}}$ against temperature for mineral oils: (a) top: B.O.C. "450" oil from 20°C. to 85°C. (b) bottom: B.O.C. "450" oil and "60" oil from 5°C. to 85°C.

is known (Chemical Engineers Handbook, 1950) that the viscosity versus temperature graphs for many liquids can be fitted to each other and to a "generalized" graph by an appropriate shift parallel to the temperature axis. This is seen to be roughly true in the present case, because a translation of the upper graph of Fig. 1(b) through about 40°C. parallel to the temperature axis will bring it into near-coincidence with the corresponding portion of the lower graph. Carrying the comparison between the two graphs a little further, we should expect another change of slope in the lower graph in the neighbourhood of $1/\nu^{\frac{1}{2}}=1.45$, *i.e.*, around a temperature of 95°C. To test this point, the viscosity measurements were extended to temperatures well above 100°C.

3. Measurements above 85°C.

For this purpose, a high-temperature thermostatic glycerol bath was designed and built in the laboratory. A diagrammatic sketch of the apparatus is shown in Fig. 2(a) and the electrical circuit for controlling the heater current is shown in Fig. 2(b). Essentially, the thermostat consists of a cylindrical glass vessel about 20 cm. diameter and 25 cm. high, contained in a rectangular wooden box for added insulation. The wooden box is provided with small plate-glass windows on opposite sides, one for providing illumination, and the other for observing the liquid levels in the viscometers. The top of the box is closed with a wooden lid that has suitably placed openings for the electrical heater, the stirrer, the control thermometer, and the viscometer holder. Two small openings are also provided for inserting two thermometers in such a way that their bulbs shall be as close as possible to the middle portion of the U-tube viscometer.

This thermostat was tested extensively from 70°C. upto 200°C., and it was found possible to maintain the temperature constant to within one or two hundredths of a degree, provided a certain amount of care was exercised in the setting of the electrical controls. This was considered to be quite satisfactory for the work in hand.

In the first instance, the viscosity of the heavy B.O.C. "450" oil was measured at various temperatures in the range of 80°C. to 145°C. The results of the measurements are given in Table 2 and the complete data from 5°C. to 145°C. are plotted in Fig. 3 as $1/\nu^{\frac{1}{2}}$ against temperature.

It is evident that the predicted change of slope at the higher temperatures does indeed occur, but the actual temperature of the transition is considerably lower than expected, being about 85°C. instead of 95°C. The direction of the bend in the graph is also as expected from analogy with the case of the lighter oil. Furthermore, the points for temperatures above 120°C. distinctly show the presence of another such bend at about 120°C. The obvious suggestion is that the viscous behaviour of the two oils studied changes fairly sharply after very definite temperature intervals, whose extent varies, but which may be estimated to be of the order of 40°C. Further elucidation of this point is reserved for the later part of the paper (section 7), in which additional and more accurate data are introduced and discussed.

TABLE 1(A).—KINEMATIC VISCOSITIES OF B.O.C. "450" OIL ABOVE ROOM TEMPERATURE.

Temperature (°C.) ..	20.3	25.0	30.0	40.0	50.0	55.0	60.0	70.0	75.0	80.0	85.0
ν (Stokes) ..	21.35	13.53	8.84	3.97	1.958	1.437	1.095	0.657	0.515	0.415	0.338
$[1/\nu]^{\frac{1}{4}}$ ..	0.465	0.521	0.580	0.708	0.845	0.913	0.930	1.111	1.180	1.246	1.313
$[1/\nu]^{\frac{1}{3}}$ ..	0.360	0.419	0.484	0.632	0.800	0.886	0.971	1.150	1.247	1.341	1.439

TABLE 1(B).—KINEMATIC VISCOSITIES OF B.O.C. "450" OIL BELOW ROOM TEMPERATURE.

Temperature (°C.) ..	15.5	9.9	5.0
ν (Stokes) ..	35.4	66.5	123.5
$[1/\nu]^{\frac{1}{4}}$ ..	0.410	0.351	0.300
$[1/\nu]^{\frac{1}{3}}$ ..	0.304	0.247	0.201

TABLE 1(C). KINEMATIC VISCOSITIES OF B.O.C. "60" OIL FROM 5 C. TO 85 C.

Temp. (°C.)	4.4	10.7	15.5	20.3	25.0	30.0	40.0	50.0	55.0	60.0	70.0	75.0	80.0	85.0
ν (Stokes) ..	3.925	2.290	1.702	1.163	0.867	0.647	0.381	0.239	0.1950	0.1638	0.1180	0.1014	0.0874	0.0756
$[1/\nu]^{\frac{1}{4}}$ ..	0.711	0.813	0.876	0.963	1.036	1.115	1.273	1.430	1.505	1.571	1.706	1.771	1.840	1.908
$[1/\nu]^{\frac{1}{3}}$ ..	0.634	0.759	0.838	0.951	1.050	1.156	1.380	1.610	1.726	1.829	2.040	2.145	2.253	2.364

TABLE 2.—KINEMATIC VISCOSITY OF B.O.C. "450" OIL AT HIGH TEMPERATURES.

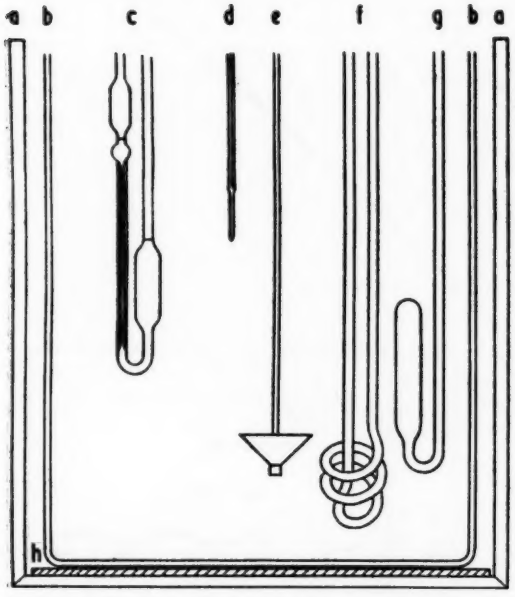
Temperature(°C.)	85	90.3	95.1	100.0	105.5	110.2	115.5	120.3	125.2	130	134.9	140	144.8
ν (Stokes) ..	0.3435	0.2836	0.2388	0.2038	0.1723	0.1485	0.1281	0.1126	0.1012	—	0.0812	—	00.658
$[1/\nu]^{\frac{1}{4}}$ ..	1.309	1.372	1.431	1.490	1.552	1.614	1.675	1.727	1.769	—	1.876	—	1.978
$[1/\nu]^{\frac{1}{3}}$ ..	1.430	1.523	1'611	1.701	1.798	1.888	1.985	2.073	2.146	—	2.308	—	2.477

TABLE 4.—"SMOOTHED" VALUES OF $\varepsilon/k \times 10^{-3}$ FOR B.O.C. "450" OIL.

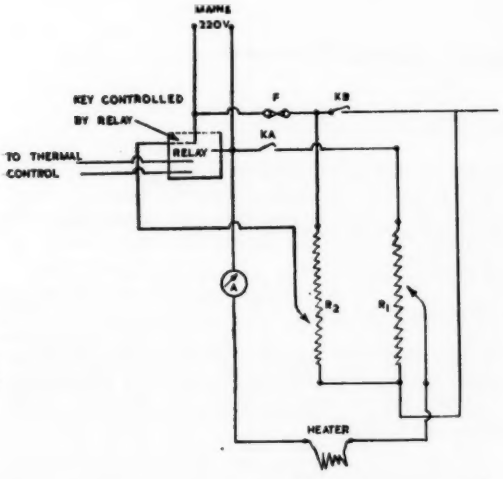
Temperature (°C.) ..	20	40	60	80	100	120	140	160	180	200	220	240
$\left(\frac{\varepsilon/k}{1000}\right)_{\text{smoothed}}$ ..	8.66	7.32	6.21	5.32	4.62	4.08	3.68	3.38	3.12	2.95	2.85	2.82

TABLE 5

Temperature (°C.) ..	30	35	40	45	50	55	60	65	70	75	80
$\frac{\varepsilon'/k}{1000}$ ..	8.79	8.09	7.57	7.11	6.73	6.43	6.17	5.94	5.74	5.57	5.42
First differences ..		0.70	0.52	0.46	0.38	0.30	0.26	0.23	0.20	0.17	0.15



a, Wooden box; b, glass vessel; c, viscometer; d, centigrade thermometer; e, stirrer; f, heater; g, thermal control; h, asbestos sheet.



R1 & R2, Reheostats; KA & KB, keys; F, fuse; A, ammeter.

Fig. 2.—Sketch of high temperature glycerol thermostat and the associated temperature control circuit.

For the time being, we analyse the practically important portions of these graphs, which fall in the viscosity range 5,000 to 10 centistokes.

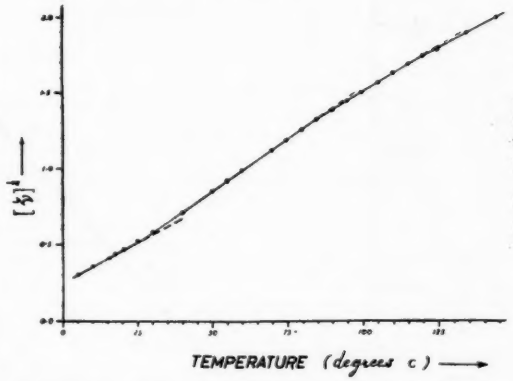


Fig. 3.—Extended graphs of $(1/\nu)^{\frac{1}{3}}$ against temperature for B.O.C. "450" oil.

4. Discussion of the $[1/\nu]^{\frac{1}{3}}$ graphs

Within the above-mentioned range, the graphs for $[1/\nu]^{\frac{1}{3}}$ consists of three straight lines, whose intersections give two quite sharp bends in opposite directions, thus giving the whole graph a sort of chair-like shape. In Fig. 4, the previously obtained data for B.O.C. "450" oil are plotted in the form of $[1/\nu]^{\frac{1}{3}}$ against temperature. Comparison of Fig. 4 with Fig. 3 brings out the fact that, although the various segments of the curves obtained can still be approximately represented by straight lines, a systematic curvature is detectable in each of the segments in Fig. 4. This curvature is best seen by separately plotting the deviation, Δ, of the observed points from the best straight lines drawn through them. These deviation plots are given in the inset to Fig. 4, and it is seen that the plot corresponding to each segment of the original graphs shows a downward convexity, which indicates that the correct index to be used for $1/\nu$ to get linearity is lower than 1/3. The exact value, n, of this index that will give linearity in any particular segment of the graph can be got by means of the following formula, which is developed in Appendix I :

$$\tfrac{1}{3}-n=\frac{\overline{y}}{3}\times\frac{\overline{d^2\Delta}}{\overline{dT^2}}\Big/\left(\frac{dy}{dT}\right)^2 \qquad (1)$$

where $y=[1/\nu]^{\frac{1}{3}}$, $\overline{y}$=mean value of y in the range considered, Δ=deviation of y from the best straight line through the points, T=temperature. With this formula, we get for the three segments of the graph for B.O.C. "450" oil, 1/3—n=0.054, 0.097, 0.081, respectively, giving a mean value of 1/3—n=0.077±0.012, whence n=0.256±0.012. 2(a)

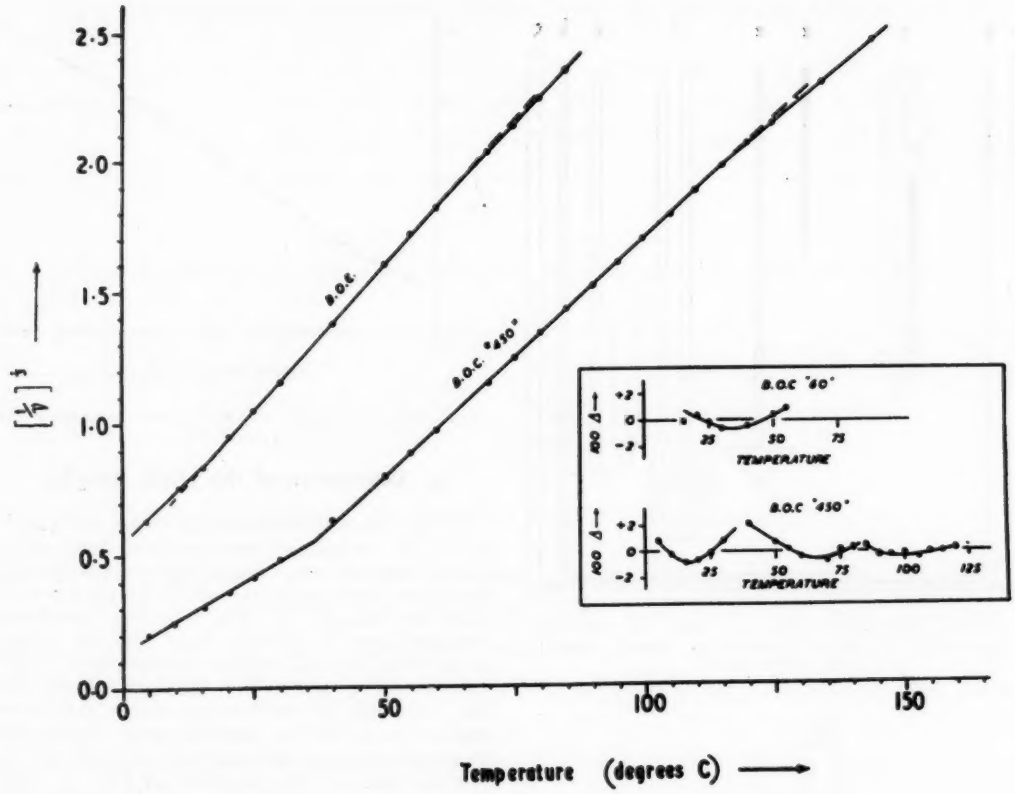


Fig. 4.—Data on B.O.C. "450" and "60" oils plotted as $(1/\nu)^{\frac{1}{3}}$ against temperature. The inset shows the deviations from the best linear segments.

The corresponding plot for the B.O.C. "60" oil gives

$$n=0.25_8 \qquad 2(b)$$

(No internal estimate of the standard deviation could be got in this case because the experimental points on the two other segments are too few to give reliable estimates for 1/3—n.) From the results 2(a) and 2(b), it follows that for both these oils the index, n, may be taken to lie definitely between 0.25 and 0.27 over the entire range of temperatures studied, *i.e.*, from 5 °C. to over 100 °C.

We may now represent the viscosity of a mineral oil by means of the formula

$$[\nu]^{-0.26}=a+bT, \qquad 3(a)$$

where a, b, will be constant in any given segment of the graphs of Fig. 3. If we further limit ourselves to the most important range of viscosities for lubricants, *i.e.*, approximately from 25 centistokes to 1,500 centistokes, we find that the constant, b, has very nearly the same value for all mineral oils, *viz.*,

$$b=0.015_2\pm0.001, \qquad 3(b)$$

In this way we obtain the following generally useful equation giving the viscosity of lubricating oils at various temperatures (in degrees centigrade or Kelvin) :

$$1/\nu^{0.26}=a+0.0152\ T, \qquad (4)$$

where the quantity, a, varies with the grade of the oil, and can be determined from equation (4) provided we measure the viscosity of the oil at any one convenient temperature. ('a' is of course to be inversely correlated with the boiling range of the oil).

The Chemical Engineers Handbook gives a widely applicable mean viscosity temperature curve. It is instructive to make a comparison between the graphs of Fig. (3) and the corresponding plots of $1/\nu^{0.26}$ against T as deduced from this universal curve (Fig. 5). The graph of Fig. 5 is very nearly linear in the middle range of viscosities, and the slope (0.0165 per °C.) is moreover very close to that given by equation (4) above, all in good agreement with the foregoing conclusions. For higher as well as for lower viscosities, the slope decreases (as in Fig. 3), but the distinction between the various segments observable in Fig. 3 is more or less completely lost in the "universal" curve of Fig. 5. This is to be expected if we remember that in different oils and other liquids the temperature for transition between segments varies somewhat and therefore the average curve will become a smooth unbroken one.

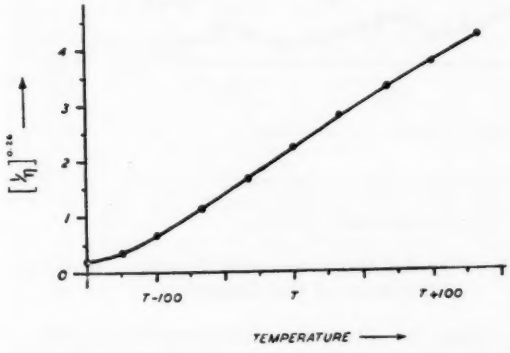


Fig. 5.—$(1/\eta)^{0.26}$ plot calculated from the generalized viscosity curve for liquids.

5. Correlation with Inter-molecular Energy

Besides their application to practical problems, the above results are interesting in themselves, and it is worthwhile to discover their fundamental connection with the phenomena occurring inside the liquids. Mitra[2] has developed the theory for a $1/T^3$ law for the variation of viscosity, based on the free-volume theory of liquids. The $1/T^3$ law was first given empirically by Batschinsky,[3] and is in fair agreement with the data on several simple liquids. On this basis, the reciprocal of the cube-root of the viscosity may be expected to vary linearly with temperature for such simple liquids ; this is very close to the law,

$$\frac{1}{\nu^{0.26}} = a + b\,T,$$

that we have found to hold for the individual segments of the graphs of Fig. 3. (It is to be noted that the kinematic viscosity ν and the dynamic viscosity η may be interchanged in this type of formula without much error, because $\nu=\eta/\rho$, and the variation in ρ is ordinarily many times smaller than that in η). Thus we may conclude that in the range corresponding to any one segment of these graphs, the oil behaves at least approximately like one of the simple non-associated liquids. The constant b, is of course dependent on the thermo-dynamic state of the liquid.

By differentiation of the foregoing viscosity temperature relation, we easily get $b = \frac{-1}{\nu^{1.26}} \frac{\partial \nu}{\partial T}$ whence b can be evaluated. In order to bring out clearly the connection between this constant 'b' and the molecular state of the liquids investigated, it is better (cf. equation 7 below) to make use of the Andrade equation, which is

$$\eta = A \exp\left(\frac{c}{T}\right) = A \exp\left(\frac{\varepsilon}{kT}\right),$$

$$\text{or } \ln \eta = \ln A + \frac{\varepsilon}{kt}, \qquad (5)$$

where "ε" has the dimensions of energy, and has been called the energy of activation of viscous flow. The general theory underlying the Andrade equation shows that the quantity ε is a measure of the mutual potential energy of the molecules or molecular aggregates in the liquid, while A can be considered as a measure of the concentration of these aggregates. For simple monatomic liquids, ε is practically constant, while for the majority of liquids it is found to vary with temperature possibly tending to a minimum near the boiling point. To a first approximation, both ε and A can be taken as constant over a small temperature range. Then by differentiation of equation (5), we get

$$\frac{\partial \ln \eta}{\partial T} = \frac{1}{\eta}\frac{\partial \eta}{\partial T} = -\frac{\varepsilon}{kT^2},$$

$$\text{whence } \frac{\varepsilon}{k} = -\frac{T^2}{\eta}\frac{\partial \eta}{\partial T} = -\frac{T^2\,\Delta \ln \eta}{\Delta T}, \qquad (6a)$$

If we neglect the small difference between $\Delta \ln \eta$ and $\Delta \ln \nu$, then this equation leads to the following connection between ε and b,

$$b = -\frac{1}{\nu^{1.26}}\frac{\partial \nu}{\partial T} = \frac{-1}{\nu^{0.26}}\frac{\partial \ln \nu}{\partial T} \simeq \frac{\varepsilon/k}{T^2 \times \nu^{0.26}} \qquad (7)$$

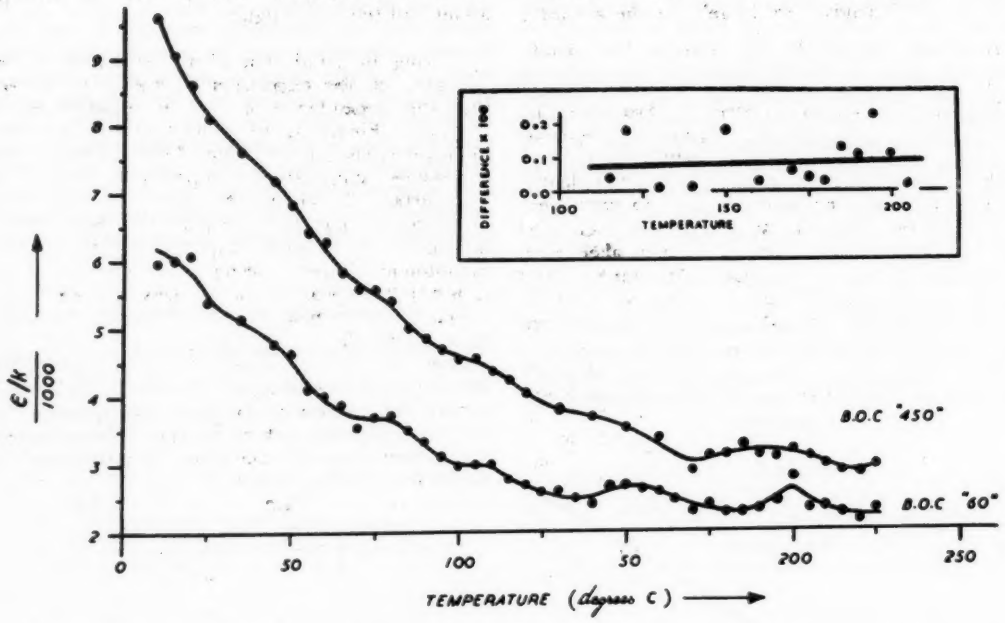


Fig. 6.—Graphs for the activation energy, ε, of the two mineral oils as a function of temperature upto 225°C. The regularly recurring segments are in evidence. The inset shows the differences between the values of $\Delta \ln t / \Delta T$ for two sets of measurements on one oil.

For $\nu < 100$ centistokes, $(T^2 \times \nu^{0.26})$ varies rather slowly, so that b is approximately proportional to ε.

When ε is constant over an appreciable range of temperatures, equation (6a) is exact, and when ε varies with temperature, this equation can be taken to give the mean value of ε/k in the temperature-range for which $\Delta \ln \eta$ and ΔT are measured. The value of A is then obtained by substitution for ε in equation (5). This gives

$$\ln A = \ln \eta - \frac{\varepsilon}{kT} = \ln \eta + T \frac{\Delta \ln \eta}{\Delta T} \qquad (6b)$$

Thus if the viscosity is measured at a number of closely spaced temperatures, it is possible to obtain from equations (6a) and (6b) values of ε and ln A over the whole experimental range of temperatures. This procedure was accordingly applied to the data already obtained on the B.O.C. "450" and "60" oils. The results (Fig. 6) were so encouraging that it was decided to extend the observations on both oils up to temperatures above 200°C. Some points in the experimental technique for these measurements deserve comment.

6. Special Experimental Features for Measurement of the Energy ε

Since the quantity to be determined accurately for equations (6a) and (6b) is $\frac{\Delta \ln \eta}{\Delta T}$ rather than η, absolute measurements of the viscosity η can now be advantageously replaced by relative measurements, so that the accuracy is not limited by the accuracy of calibration of the U-tube viscometer. Furthermore, it is not essential to adjust the level of the liquid in the viscometer for each temperature. It is much better to leave the experimental liquid entirely enclosed (and safe from contamination) for a complete set of observations extending over about 40°C., and to apply an appropriate correction for the changing level. This level correction is small, being of the order of 0.1% for one mm. rise in level, which corresponds to a rise in temperatures of about 3°C. The correction for changing kinematic energy of flow (through the capillary) is also small, and is readily applied. The calculations for these and other corrections are worked out in Appendix II.

Since $\frac{\Delta \ln \eta}{\Delta T} \simeq \frac{\Delta \ln t}{\Delta T}$, where t is the time of

flow at temperature T, the accuracy of the determination of ε depends very largely on the accuracy with which $\Delta \ln t = \ln \frac{t_2}{t_1}$ and $\Delta T = T_2 - T_1$ can be measured. In order to increase the accuracy of the measurements of flow-time, the observation at any one temperature was repeated four to five times, the temperature being kept constant to within a few hundredth of a degree centigrade. Observations of the flow-time were made at intervals of approximately 5°C., and the values for each temperature were taken twice first during the rising temperature sequence and then with temperature fal ing. In this way, the time of flow at each temperature was obtained accurate to better than 1 part in 2,000, so that the values of $\Delta \ln t$, *i.e.*, $\ln t_2/t_1$ calculated from these measurements are accurate to at least 0.001. Therefore, if we aim at an accuracy of 2% in the determination of $\Delta \ln t/\Delta T$, we must adjust ΔT so that $\Delta \ln t$ is of the order of 0.1. This condition requires ΔT to be of the order of 10°C. in the present case. The value of ΔT should also be measured correct to 1%, and therefore the temperature differences must be obtained accurate to 0.1°C., which can be accomplished with a half-degree or even a degree thermometer equipped with a magnifying thermometer-reader. Because of the foregoing consideration, a 10°C. interval was used for ΔT in the calculations, although most of the observations were taken every 5°C. This procedure will of course mask some of the small-scale variations in ε that occur within a temperature range of 10°C.

7. Discussion of the Graphs for ε against Temperature

The results obtained for B.O.C. "60" and "450" oils are given in Table 3, and the quantity $\frac{\varepsilon}{k} = -T^2 \Delta \ln \nu / \Delta T + T^2 \times \beta$ is plotted against temperature in Fig. 6. The inset to this figure shows a plot of typical differences between the values of $-\frac{\Delta \ln t}{\Delta T}$ for the sets of observations taken with rising and falling temperature sequences: it is seen that, the mean difference is of the order of 0.8×10^{-3} so that the standard deviation of the mean is nearly 0.5×10^{-3}, which corresponds to an error of 0.05 to 0.1 in the value of $\frac{\varepsilon/1000}{k}$. This is as expected from the foregoing analysis of the experimental technique. It is to be noted that since each small portion of the graph for $\frac{\varepsilon/k}{1000}$ against temperature is drawn as a sort of mean of the two or three nearest experimental points, the error in the graph will be even less perhaps about half the above figure.

Bearing in mind the above estimates of the accuracy of the experimental points, it is seen that the dependence of ε/k on temperature is two-fold. Firstly, there is the well-known monotonous decrease in the value of this quantity with increasing temperature; superposed upon this is a further sinusoidal variation with a cycle of about 30°C. to 40°C., which becomes particularly prominent at higher temperatures where the monotonous decrease becomes small. The graph thus has the appearance of a succession of segments, which change somewhat gradually from one to the next. This fully confirms the occurrence of "knees" or "bends" observed in the earlier plots of $[1/\nu]^{1/4}$ against temperature, and points to the occurrence of certain changes in the arrangement of the molecules within the oil. (The "knees" correspond to the portions joining successive segments with downward convexity. Further analysis of these segments is given later on in the next section). Here it is desirable to examine the form of the general average variation of ε/k with temp ture. For instance, if through the observed points for B.O.C. "450" oil, a mean curve is drawn ignoring the sinusoidal variation, this curve is found to be approximately parabolic. (The inclusion of small terms in T^3 and T^4 gives a virtually perfect fit). The ordinates for this "smoothed" curve are given in Table 4 at intervals of 20°C. (cf.p. 118). A least-squares treatment gives the following best parabolic equation for this curve :—

$$\left(\frac{\varepsilon/k}{1000}\right)_{\text{smoothed}} = 2.80 + 1.4\left(\frac{T-493}{100}\right)^2 \pm 3\%$$

(T in degrees Kelvin)

The differences between this equation and the values of Table 4 are small, and lie within the superposed sinusoidal variations in the experimental curves of Fig. 6. Similar results are got with B.O.C. "60" oil, so that we may take, as a working approximation for these oils, the general equation,

$$\frac{\varepsilon}{k} = -T^2 \frac{\Delta \ln \eta}{\Delta T} = c + b\,(T_o - T)^2, \qquad (8)$$

$$i.e., \quad \frac{\Delta \ln \eta}{\Delta T} = -\frac{c + b\,T_o^2}{T^2} + \frac{2b\,T_o}{T} - b,$$

which can be integrated to give

$$\ln \eta = \frac{c + bT_o^2}{T} + 2b\,T_o \ln \frac{T}{T_o} - bT + \ln A_o$$

TABLE 3.—VALUE OF $\frac{\varepsilon/k}{1000} = -\frac{T^2}{1000}\frac{\Delta \ln \nu}{\Delta T} + \frac{T^2\beta}{1000}$ FOR VARIOUS TEMPERATURES BETWEEN 10°C AND 225°C.

(β varies from 0.000,55 to 0.000,75) —B. O. C. "450" Oil—						(β varies from 0.000,60 to 0.000,95) —B. O. C. "60" Oil—					
Mean Temp.	$-\Delta\ln t/\Delta T$ Temp. rising	Temp. falling	Mean	$-\frac{T^2}{1000}\frac{\Delta\ln\nu}{\Delta T}$	Correction $=\frac{T^2\beta}{1000}$	Mean Temp.	$-\Delta\ln t/\Delta T$ Temp. rising	Temp. falling	Mean	$-\frac{T^2}{1000}\frac{\Delta\ln\nu}{\Delta T}$	Correction $=\frac{T^2\beta}{1000}$
10				9.64	0.04	10				5.91	0.05
15				8.99		15				5.96	
20				8.55		20				6.02	
25	Calculated from previous data			8.05	0.05	25	Calculated from previous data			5.32	0.06
30	of Table 1.			—		30	of Table 1.			—	
35				7.55		35				5.08	
40				—		40				—	
45				7.13		45				4.70	
50				6.76	0.06	50				4.56	0.07
55				6.36		55				4.04	
60				6.20		60				3.95	
65				5.76		65				3.80	
70				5.51		70				3.47	
75				5.50	0.08	75				3.61	0.09
80				5.33		80				3.65	
85		—	.03845	4.93		85	.0267	.0266	.02665	3.42	
90	—	—	.03615	4.76		90	.0251	.0241	.0246	3.24	
95	—	—	.0338	4.59		95	.0218	.0229	.02235	3.02	
100	—	—	.0320	4.45	0.09	100	.0204	.0209	.02065	2.87	0.11
105	—	—	.0312	4.46		105	.0210	.0195	.02025	2.89	
110	—	—	.02915	4.27		110	.0198	.0197	.01975	2.90	
115	.0272	.0276	.0274	4.13		115	.0171	.0184	.01775	2.67	
120	.0245	.0263	.0254	3.93		120	.0160	.0174	.0167	2.58	
125	—	—	—	—	0.11	125	—	.0156	.0156	2.47	0.13
130	0.227	.0226	.02265	3.68		130	.0158	.0148	.0153	2.48	
135	—	—	—	—		135	—	.0142	.0142	2.37	
140	.0212	.0211	.02115	3.60		140	.0132	.0133	.01325	2.26	
145	—	—	—	—		145	—	.0145	.0145	2.53	
150	.0200	.0181	.01905	3.41	0.13	150	.0142	.0142	.0142	2.54	0.15
155	—	—	—	—		155	—	.0136	.0136	2.48	
160	.0173	.0176	.01745	3.27		160	.0131	.0128	.01295	2.43	
165	—	—	—	—		165	—	.0120	.0120	2.31	
170	.0143	.0137	.0140	2.76		170	.0106	.0111	.01085	2.13	
175	.0150	.0146	.0148	2.97	0.15	175	—	.0111	.0111	2.22	0.18
180	.0147	.0144	.01455	2.99		180	.0105	.0099	.0102	2.09	
185	.0155	.0142	.01485	3.12		185	—	.0100	.0100	2.09	
190	.0133	.0144	.01385	2.97		190	.0109	.0091	.0100	2.14	
195	.0145	.0122	.01335	2.93		195	—	.0103	.0103	2.25	
200	.0141	.0130	.01355	3.03	0.17	200	—	.0116	.0116	2.60	0.21
205	.0127	.0129	.0128	2.93		205	—	.0092	.0092	2.10	
210	.0120	—	.0120	2.80		210	—	.0092	.0092	2.14	
215	.0113	—	.0113	2.69		215	—	.0086	.0086	2.04	
220	.0110	—	.0110	2.67		220	—	.0079	.0079	1.92	
225	.0111	—	.0111	2.75	0.20	225	—	.0083	.0083	2.06	0.25

NOTE :—The oils gave evidence of some chemical changes on heating above 200°C. as shown by a deepening of the colour. Therefore, the last five sets of readings have only a limited significance.

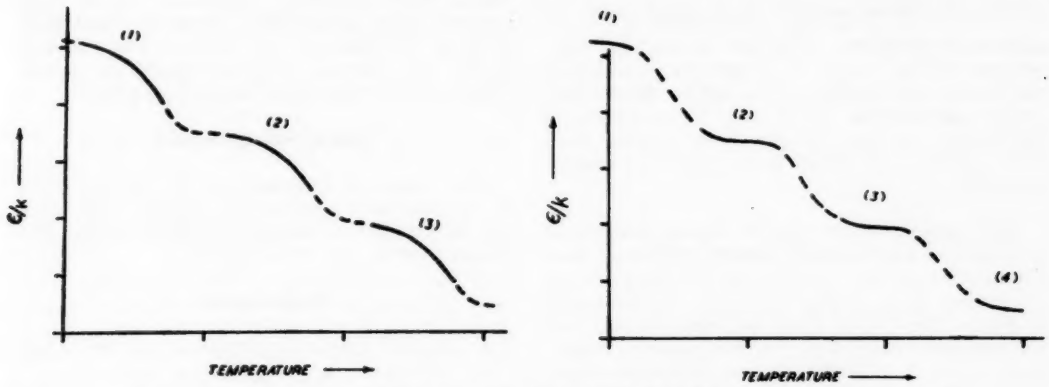


Fig. 7.—Schematic representation of two possible modes of visualization of the segments composing the curves for activation energy against temperature.

($\ln A_0$ = constant of integration)

$$= (c+b(T_0-T)^2)/T+2b\,(T_0 \ln \frac{T}{T_0}-T+T_0)+ \ln A_0$$

$$= \frac{\varepsilon}{kT}+2b\left[\, T_0\left(\frac{T-T_0}{T_0}-\frac{1}{2}\left(\frac{T-T_0}{T_0}\right)^2 +\frac{1}{3}\left(\frac{T-T_0}{T_0}\right)^3 \ldots\ldots\right)+(T_0-T)\,\right]+\ln A_0$$

$$= \frac{\varepsilon}{kT}+\ln A_0-\frac{b}{T_0}(T_0-T)^2\,(1+\frac{2}{3}\frac{T_0-T}{T_0}+..) \quad (9)$$

Comparison of this result with the logarithm of the Andrade equation

$$\ln \eta = \ln A + \frac{\varepsilon}{kT}$$

gives us

$$\ln \frac{1}{A} = -\ln A \simeq -\ln Ao + \frac{b}{T_0}(T_0-T)^2 \quad (10)$$

thus showing that the variation of $\ln 1/A$ runs more or less parallel to that of $\frac{\varepsilon/k}{T_0}$.

This result is illuminating, because it shows how the variation of the viscosity with temperature can at least in theory be decomposed into its two elements of

(1) a nearly parabolic decrease of the potential energy, ε, with increase of temperature, and

(2) a corresponding increase in ln A, where A may be taken to indicate the number of molecular aggregates taking part in the viscous interaction.

Since A increases, it follows that the aggregates become smaller at higher temperatures, which is surely reasonable, because the degree of association is then known to decrease. It is also reasonable to expect that as the size of the aggregates become smaller, so will the mutual potential energy tend to decrease. This decreasing mutual potential energy (at contact) is of course to be correlated with two factors (a) the decrease in the strength or number *per aggregate* of the *exposed* polar (or other type of bonding) groups present in the molecules, and (b) the increase in vibration and rotations of the atomic groups forming the molecular aggregates. These two factors may to some extent be interdependent.

8. Conclusions

Bearing these points in mind, we now examine the approximately sinusoidal fluctuation present in the curves for the potential energy, ε. The curves can be split up into segments, each of which consists of a steep portion and a comparatively flat portion, so that we may visualize the whole curve as being built up in one of two possible ways :—

(a) A series of parabolic segments, convex upwards, as shown schematically in Fig. 7 (a), with a more or less smooth transition from one to the next.

(b) The other alternative is to consider the curve ε against temperature to be made up of parabolic segments, concave upwards, again with a smooth transition between successive segments.

It is shown in Appendix III that each one of these particular parabolic segments now corres-

ponds to the linear segments of the graphs for $\frac{1}{v^{2.26}}$ against temperature. This can be seen by comparison of Fig. 6 with the temperatures at which the bends occur in Fig. 3, *viz.* 28°C., 86°C., and 120°C. for B.O.C. "450" oil. (The graphs of Fig. 6 give a hint that the long segment from 28°C. to 80°C. is in fact made up of two shorter segments).

This second scheme can be further elaborated to consist of short approximately horizontal lines joined together by the S-shaped sections shown by broken lines in the schematic representation of Fig. 7 (b). This mode of representation has the two advantages that the flat portions correspond to an ideal liquid obeying Andrade's equation (with constant A and ε), and the transition from one such state to the next (as shown by the broken lines) would correspond to a change of the second-order, somewhat along the lines of order $\rightleftharpoons$ disorder transitions in alloys.

Thus, on fundamental grounds, and because of the linear v-$0.^{26}$ plots, the second mode of visualization is to be preferred. We shall accordingly adopt this as an aid to further study, without in any way prejudicing the question of the precise mechanism responsible for the phenomena being investigated. Viewed in this light, the curves of Fig. 6 and 7(b) raise the following important points :—

(1) What is the significance of the energy differences between the various "steps" or segments in Fig. 7(b) ?

(2) In what way does this energy difference depend on the temperature ?

(3) What precise meaning is to be attached to the quantity A, which we provisionally label as the "Concentration factor?"

(4) What simple postulates can give a comprehensive theory describing the temperature dependence of viscosity *in full detail* ?

Since it is known that mineral oils are not pure liquids with a unique chemical formula, but are mixtures of several related hydrocarbons, the data on these oils can at this stage only point the way for further work. They also indicate the need for measurements of considerably greater precision than has been attained in the foregoing set of experiments. In order to get definite answers to the questions just raised, we have undertaken a series of investigations on several pure liquids and solutions, especially those with exposed polar groups like the mono-, and poly-alcohols, the phenols, and water. These investigations are already yielding significant results, which will be reported in succeeding papers.

Acknowledgements

The author is indebted to Mr. Salim Malik for helping with much of the experimental work, and to Dr. Salimuzzaman Siddiqui for his continued interest and encouragement

References

1. M.M. Qurashi, Pakistan J. Sci. Ind. Research, **1**, 7 (1958).
2. S.S. Mitra, J. Indian Chem. Soc. **32**, 297 (1955).
3. Batschinsky, Bull. Soc. Natur. Moscow, **15**, 1 (1901).
4. E.N. da C. Andrade, Phil. Mag., **17**, 497, 698 (1934).

Appendix I

Estimation of Index n

Suppose that $1/v^n = a + bT$, where n is the correct value of the index, and that
$y = 1/v^{n'} = a' + b'T + \Delta$
where n' is an approximate trial value of the index, and a', b' are the constants in the linear equation that gives the best fit in this case. Differentiation of these two equations then gives

$$\frac{d}{dT}(v^{-n}) = -nv^{-n-1}\frac{\partial v}{\partial T} = b, \text{ } i.e., \frac{\partial v}{\partial T} = -\frac{b}{n}v^{n+1},$$

and

$$\frac{dy}{dT} = \frac{d}{dT}(v^{-n'}) = -n'v^{-n'-1}\frac{dv}{dT} = -n'v^{-n'-1} \times -\frac{b}{n}v^{n+1} = \frac{n'}{n}bv^{n-n'}.$$

Further differentiation gives

$$\frac{d^2y}{dT^2} = b\frac{n'}{n}(n-n')v^{n-n'-1}\frac{\partial v}{\partial T} = b\frac{n'}{n}(n-n')v^{n-n'-1} \times \left(-\frac{b}{n}\right)v^{n+1}$$

$$= b^2\frac{n'-n}{n}\times\frac{n'}{n}\times v^{2n-n'}$$

$$= \frac{n'-n}{n'}\times b^2\times\left(\frac{n'}{n}\right)^2\times(v^{n-n'})^2v^{n'}$$

$$= \frac{n'-n}{n'}v^{n'}\left(-\frac{dy}{dT}\right)^2.$$

$$\text{Thus } n'-n = n' \times \nu^{-n'} \times \frac{d^2y}{dT^2} \Big/ \left(\frac{dy^2}{dT}\right)$$

$$= n' \times y \times \frac{d^2\Delta}{dT^2} \Big/ \left(\frac{dy}{dT}\right)^2$$

$$= n' \times \left(y \frac{dT}{dy}\right)\left(\frac{d^2\Delta}{dT^2} \Big/ \frac{dy}{dT}\right) \qquad (11)$$

since $y = 1/\nu^{n'}$

and $\dfrac{d^2y}{dT^2} = \dfrac{d}{dT^2}(a'+b'T+\Delta) = \dfrac{d^2\Delta}{dT^2}$.

Now, the mean value of $d^2\Delta/dT^2$ for any one segment can readily be obtained from the curvature of the corresponding deviation plot for Δ against T ; the corresponding value of y to be used is the mean for that segment.

Appendix II

Viscometer Corrections

At temperature T_1 the level is x_1 mm. above the mark, the mean time of flow being t_1 seconds, and at temperature T_2 the level is x_2 mm. above the mark, the mean time of flow being t_2 seconds. If the flow-time is increased by α % for every 1 mm. rise in level ($\alpha \sim$ 0.1), then the corrected times of flow at the two temperatures are

$$(t_1)_{corr} = t_1\left(1 - \frac{\alpha x_1}{100}\right)$$

and $$(t_2)_{corr} = t_2\left(1 - \frac{\alpha x_2}{100}\right).$$

The standard formula for the U-tube viscometers is

$$\nu = A\,t - B/t = A\,t\left(1 - \frac{B/A}{t^2}\right)$$

whence $$\ln \nu = \ln (A\,t) + \ln\left(1 - \frac{B/A}{t^2}\right)$$

$$= \ln (A\,t) - \frac{B/A,}{t^2}$$

to the first order of small quantities. Using this equation, we get to a sufficient accuracy,

$$\ln \nu_1 = \ln\left[A\,t_1\left(1 - \frac{\alpha x_1}{100}\right)\right] - \frac{B/A}{t_1^2}$$

$$= \ln (At_1) - \frac{\alpha x_1}{100} - \frac{B/A,}{t_1^2}$$

and $$\ln \nu_2 = \ln\left[At_2\left(1 - \frac{\alpha x_2}{100}\right)\right] - \frac{B/A}{t_2^2}$$

$$= \ln(At_2) - \frac{\alpha x_2}{100} - \frac{B/A,}{t_2^2}$$

whence $$\Delta \ln \nu = \ln \nu_2 - \ln \nu_1 = \ln \frac{t_2}{t_1}$$

$$- \frac{\alpha}{100}(x_2 - x_1) - \frac{B}{A}\left(\frac{t_1^2 - t_2^2}{t_1^2 \times t_2^2}\right)$$

$$= \ln \frac{t_2}{t_1} - \frac{\alpha}{100}(x_2 - x_1) + 2\frac{B/A}{t^2}\frac{t_2 - t_1}{t}$$

where $t = \frac{1}{2}(t_1 + t_2)$. It follows that

$$\frac{\Delta \ln \nu}{\Delta T} = \frac{\ln t_2/t_1}{T_2 - T_1} - \frac{\alpha}{100}\frac{x_2 - x_1}{T_2 - T_1}$$

$$+ 2\frac{B/A}{t^2}\left\{\frac{(t_2 - t_1)/t}{T_2 - T_1}\right\}$$

$$= \frac{\ln t_2/t_1}{T_2 - T_1}\left(1 + 2\frac{B/A}{t^2}\right) - \frac{\alpha}{100}\frac{x_2 - x_1}{T_2 - T_1}, \qquad (12)$$

because the term in curly brackets is very nearly equal to $\left(\ln \frac{t_2}{t_1}\right)/(T_2 - T_1)$. In a typical case, $B/A \sim 40$, t is of the order of 200 secs., so that the kinetic energy correction term amounts to about $80/(200)^2$, *i.e.*, 0.2%. When $(T_2 - T_1)$ is 5 °C., $\ln \frac{t_2}{t_1}$ is of the order of 0.1 and $x_2 - x_1$ is of the order of 2 mm., so that both the corrections together amount to about 1% of the measured quantity, $\Delta \ln \nu/\Delta T$. Another correction should be applied for the change in viscometer constant due to its thermal expansion, but this can be shown to be negligibly small, because the coefficient of cubical expansion of the glass is of the order of 0.000,02. Finally we must note that, for equations (6a) and (6b), we require $\Delta \ln \eta/\Delta T$, whereas experimentally we obtain $\Delta \ln \nu/\Delta T$ from equation (12) above. The difference between these quantities is given by

$$\frac{\Delta \ln \eta}{\Delta T} = \frac{\Delta \ln (\nu \times \rho)}{\Delta T} = \frac{\Delta \ln \nu}{\Delta T} + \frac{1}{\rho}\frac{\partial \rho}{\partial T}$$

$$= \frac{\Delta \ln \nu}{\Delta T} - \beta, \qquad (13)$$

where β is the coefficient of cubical expansion of the liquid being investigated. Since the present investigation is confined to temperatures below the boiling point, β is more or less constant, and

will be of the order of 0.000,5. Since $\Delta \ln \nu/\Delta T$ is of the order of 0.02, the error introduced by neglecting β in the above equation would be more than 1%, and cannot therefore be ignored. However, this correction has only a slow variation with temperature, and is readily applied by means of equation (13).

Appendix III

Correlation between the Activation Energy Graph and the $1/\nu^{0.26}$ linear Plots

Essentially, the problem is to find the variation of ε'/k corresponding to a linear plot of $1/\nu^{0.26}$ against temperature. Consider, for instance, the long second segment of the graph for B.O.C. "450" oil in Fig. 3. Its equation is

$$1/\nu^{0.25} = 0.16 + 0.0136\ (T - To),$$

differentiation of which gives

$$-0.25\ \nu^{-1.25}\frac{\partial \nu}{\partial T} = 0.0136,$$

whence $$\frac{\varepsilon}{k} \simeq -\frac{T^2}{\nu}\frac{\partial \nu}{\partial T} = T^2 \times \frac{0.0136/0.25}{\nu^{-0.25}}$$

$$= T^2 \times \frac{0.0544}{0.16 + 0.0136(T - To)}$$

Table 5 on page 118 gives the values of $\frac{\varepsilon/k}{1000}$ calculated from this relation over the range 30°C. to 80°C., together with the first differences. It is evident that the corresponding graph of $\frac{\varepsilon/k}{1000}$ against temperature will have a pronounced downward convexity, which is in agreement with the second mode of analysis of the ε/k curves into segments.

PRELIMINARY STRUCTURAL STUDY OF THE MAGNETIC FRACTION OF STEAM-TREATED MAKERWAL COAL

S. JAMAL QUADIR AND SADRUL HASAN RIZVI

Central Laboratories, Pakistan Council of Scientific and Industrial Research, Karachi

Introduction

In the course of their studies in the desulphurization of Makerwal and other sulphuraceous coals of West Pakistan by treatment with super-heated steam, Siddiqui et al.[1] noted that the beneficiated coal residue contains 5 to 10% of a strongly magnetic component. This component could be removed by simple magnetic separation, and it was found that it carried away a considerable fraction of the residual sulphur, thus providing a basis for additional beneficiation of the coal. The present communication deals with an account of a preliminary investigation in the composition and structure of this magnetic fraction.

Two Possible Paramagnetic Compounds

Since it was known that the original coal contained both sulphur and iron, all the iron being in the form of pyrites, it was at first thought that the magnetic property was due to the formation of compounds of the series Fe_nS_{n+1}, which are known to be strongly paramagnetic. Non-stoichiometric compounds of iron and sulphur, both ferromagnetic and paramagnetic, are known to occur naturally and have even been prepared synthetically ; but they form a somewhat peculiar class of compounds and their structure and magnetic behaviour are not fully understood. FeS and FeS_2 are the two well-established compounds of iron with sulphur, but they are both non-magnetic and have structures of the nickel-arsenide and pyritic types, respectively. The compounds, $Fe_{0.48}S_{0.52}$ to $Fe_{0.466}S_{0.534}$ (corresponding to $Fe_{12}S_{13}$ to Fe_7S_8 approximately), have been reported as being ferromagnetic in character, with a nickel-arsenide structure, whereas the compounds in the composition range between $Fe_{0.50}S_{0.50}$ and $Fe_{0.48}S_{0.52}$ (corresponding to FeS to $Fe_{12}S_{13}$ approx.) are reported to be

paramagnetic, again with a nickel-arsenide structure in the high sulphur half of the field but with a superstructure in the region near the ideal composition FeS. M.J. Buerger's[2] work, however, throws doubt on the reported structures and it is not safe to say anything about the structure of Fe_nS_{n+1}, except that the reported structural studies indicate that the lattice of Fe_nS_{n+1} is substantially different from that of FeS_2. So the conversion of FeS_2 into Fe_nS_{n+1} through any reaction of the type

$$FeS_2 \text{ (coal)} + HOH \text{ (Steam)} \xrightarrow{\text{at about } 300\,^{\circ}\text{C.}} Fe_nS_{n+1} + H_2S$$

would be associated with a rather improbable structural transition.

The other simple compound of iron that has a high value of magnetic susceptibility is Fe_3O_4. However, the production of Fe_3O_4 by simple superheated steam-treatment also seems somewhat unlikely, because the conversion of FeS_2 into Fe_3O_4 is known to occur only at a temperature round about 500 °C., whereas the magnetic fraction under study was reported to have been obtained at a much lower temperature (around 350 °C.).[1] Thus we have to decide between two possibilities, both of them apparently unlikely. This anomalous situation led to the present detailed study of the problem.

Chemical Analyses

To begin with, an effective separation of the magnetic component from the adhering coal was attempted. It was done initially by the ordinary dry method with a magnet, but because of the "carrying-down" tendency of coal the separation had to be carried out by the wet gravity method, followed by a further antigravity separation in the dry state by playing a magnet over a 250 mesh sieve. A magnetic fraction of reasonable "purity" was thus obtained. Preliminary chemical analyses of this fraction indicated that it does indeed contain sulphur and iron in large quantities with a comparatively small percentage of carbon. But this did not account for the total weight of the magnetic component, and there appeared to be a considerable quantity of oxygen in chemical combination. Two typical analyses of samples of the magnetic fraction are given in Table 1 together with the mean of half a dozen analyses.

From the above analyses it was clear that, compared with sulphur, the oxygen constitutes the major component of the magnetic fraction. Ignoring carbon as a mechanical impurity, the following approximate empirical formula could be derived from the above analyses :

$Fe_1O_{1\cdot38}S_{0\cdot25}$, *i.e.* $(Fe_4O_{5\cdot5}S)$ or $(Fe_3O_{4\cdot1}S_{0\cdot8})$, corresponding to (1) $Fe_2(O,S)_3$ or (2) $F_3O_4 + S$.

From this, it appeared that the magnetic component is essentially an iron oxide with a small quantity of sulphur, either incorporated in the lattice or else adsorbed. The fact that the empirical formula approximates to Fe_3O_4, (the magnetic oxide), taken together with the high magnetizability of the material, gave considerable support to this idea.

There remained now the problem of the precise status of the sulphur in the iron oxide (probably Fe_3O_4) lattice comprising the magnetic fraction.

In order to examine this, repeated extractions of the sulphur were attempted in a Soxhlet ap-

TABLE 1.—TYPICAL ANALYSES OF THE MAGNETIC FRACTION.

(Samples oven-dried at 105 °C.)

	Analysis I (weight %)	Analysis II (weight %)	Mean of six analyses (weight %)	Mean weight % / Atomic weight
Iron	53·9	58·9	56	1.00
Sulphur	5.1	8.2	8	0.25
Carbon (by combustion) ...	41.0	13.0	14	1.17
Oxygen (by difference) ..		19·9	22	1.38

paratus, using carbon tetrachloride as the solvent. It was found that each extraction removed successively smaller quantities of the sulphur, as indicated in Table 2.

TABLE 2.—SUCCESSIVE EXTRACTIONS OF SULPHUR

(Initial Sulphur Content=8%)

Serial No. of extraction	1	2	3
Sulphur removed in this extraction (weight % of initial sample)	0.28	0.12	0.10
Total sulphur removed	0.28%	0.40%	0.50%

From this table, it may be estimated by extrapolation that, even after a very large number of such extractions, a fair quantity of sulphur (about 4%) would remain in the magnetic fraction. The conclusion suggested by this is that a portion of the sulphur is present in a non-extractable form, while the rest is only mechanically bound. However considering the fact that adsorbed sulphur is frequently very difficult to dislodge by extraction, further work for clarifying this point was taken up by X-ray diffraction, which presents perhaps the simplest way of determining whether or not the sulphur is chemically combined.

X-ray Diffraction Analysis

For this analysis, it was necessary to take X-ray powder patterns of the magnetic component and to compare them with those of (i) FeS (ii) Fe_nS_{n+1} (iii) Fe_3O_4 (iv) Fe_2O_3, all of which can be suspected to contribute to the structure. Reproductions of these patterns are shown in Fig. 1, from which it is quite clear that the pattern of the magnetic fraction agrees only with that of Fe_3O_4. However, there is one noticeable difference in that there is an extra line of low intensity at d=2.71 A in our standard pattern of Fe_3O_4. This line coincides with the strongest line of the α-Fe_2O_3 pattern (Table 3), and could be due to the well-known contamination of F_3O_4 with small quantities of Fe_2O_3. By visual estimation of relative intensities, it was found that the Fe_3O_4 sample prepared by us contained approximately 10% Fe_2O_3, which is a reasonable figure. Another possible explanation of the extra line could be that it is due to the presence of some γ-Fe_2O_3, the remaining lines of whose pattern are practically indistinguishable from those of Fe_3O_4, as can be seen in Table 3.

It follows from the above data that the magnetic fraction of the treated coal consists essentially of Fe_3O_4, with a possibility of some γ-Fe_2O_3, but no detectable α-Fe_2O_3.

Conclusion

In the light of the foregoing observations, it may be concluded that the bulk of the pyritic material present in Makerwal coal is converted into Fe_3O_4 by the previously reported process of high temperature steam treatment. The different stages of the reaction FeS_2+H_2O (steam) $\rightarrow Fe_3O_4$ are given in the literature,[3] and are reported to proceed as follows :—

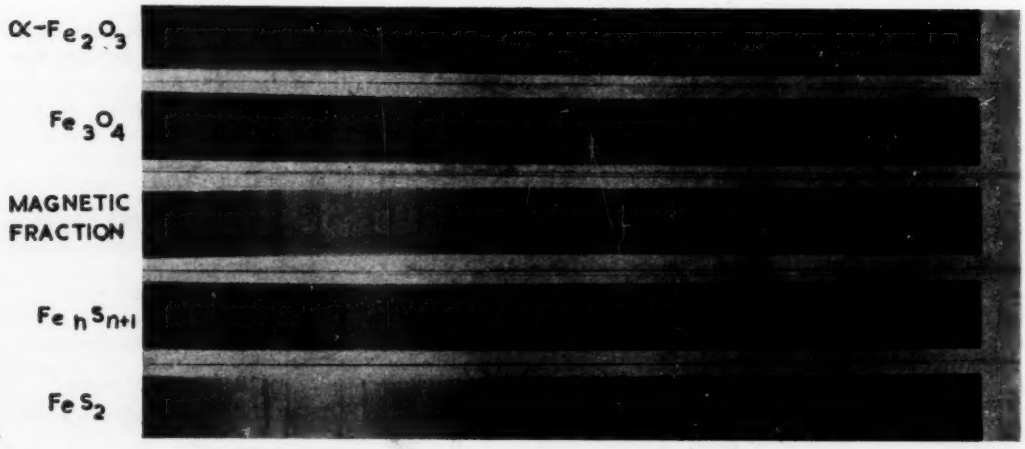


Fig. 1.—Comparative X-ray patterns for the structural analysis of the magnetic fraction. (9 cm. powder Camera, Cu K α radiation)

TABLE 3.—THE PROMINENT POWDER LINES (HAVING INTENSITIES (I/I_0) GREATER THAN 10%) OF : (1) α-Fe_2O_3, (2) γ-Fe_2O_3, (3) Fe_3O_4 AND (4) THE MAGNETIC FRACTION.

(The figures in brackets denote the intensities of the lines)

Lines of α-Fe_2O_3 (Angstroms)	Lines of γ-Fe_2O_3 (Angstroms)	Lines of Fe_3O_4 (Angstroms)	Observed lines in the magnetic fraction (Angstroms)
3.68(70)	2.95(34)	2.97(28)	2.97(30)
2.69(100)	2.78(19)		
2.51(80)	2.52(100)	2.53(100)	2.53(100)
2.20(70)		2.42(11)	
2.07(10)	2.08(24)	2.10(32)	2.10(20)
1.837(70)	1.70(12)	1.71(16)	1.70(20)
1.691(80)	1.61(33)	1.61(64)	1.61(40)
1.634(10)	1.48(53)	1.48(80)	1.48(60)
1.596(40)	1.27(11)	1.28(20)	
1.484(70)		1.12(10)	
1.451(80)	1.09(19)	1.09(32)	1.095(10)
1.348(20)		1.05(10)	1.047(10)
1.309(40)			
1.255(30)			
1.224(10)			
1.205(10)			
1.187(30)			
1.160(30)			
1.137(40)			
1.100(40)			
1.053(50)			

$$FeS_2 \rightarrow FeS + S$$

$$FeS + HOH \rightarrow FeO + H_2S$$

(cf. large quantities of H_2S that are given off in our desulphurization process).

$$2FeO + HOH \rightarrow Fe_2O_3 + H_2$$

$$3FeS + 4HOH \rightarrow Fe_3O_4 + 3H_2S + H_2$$

The occurrence of these reactions at a temperature of 350 °C. instead of the reported figure of 500 °C. may perhaps be attributed to the catalytic action of some other constituent of the coal.

There remains the possibility of the formation of small quantities of Fe_nS_{n+1} and γ-Fe_2O_3 in addition to Fe_3O_4. On the basis of the empirical formulae deduced earlier, *viz.*

$$Fe_3O_4 + S, \text{ or } Fe_2(O,S)_3,$$

it appears highly probable that the compound formed is ferroso-ferric oxide. The pronounced ferromagnetism of the material does not however preclude the possibility of some γ-Fe_2O_3, but the possibility of its formation in large quantities is ruled out by the fact that the diffraction patterns of γ-Fe_2O_3 and Fe_3O_4 do show certain differences (Table 3), and no trace of the 2.78A line of γ-Fe_2O_3 is observed in the pattern of the magnetic fraction (Fig. 1). The complete absence of Fe_nS_{n+1} also cannot be claimed with any certainty as its presence to the extent of 5% could pass undetected because of the limitations of the available techniques. With improved methods, it is hoped subsequently to present a fuller description of the status of the sulphur contained in the magnetic fraction.

References

1. Siddiqui et al., Proc. 6th Pakistan Sci. Conf. Abstr. Chem. Sec. p. 52, 1954.
2. M.J. Buerger, Am. Mineralogist, **32**, 411 (1947).
3. E. Von Grunert, J. prakt. Chem. (N.S.), **122**, 1 (1929).

PRELIMINARY ULTRA-VIOLET ABSORPTION STUDY OF THE HALOGEN ACID SALTS OF ALKALOIDS OF THE CONESSINE SERIES

A.J. Hamdani, Noorul Haq Khan, and the late S. Mehdi Ahsan

Central Laboratories, Pakistan Council of Scientific and Industrial Research, Karachi

Introduction

On the basis of studies in the von Braun[1] reaction of BrCN on tertiary bases, Siddiqui et al. [2,3] had forwarded the view that in case of conessine and isoconessine the intensity and character of the pharmacological action of conessine was very largely dependant on the direction of the change occasioned by the alteration of the N-stability of the radicals attached to the basic nitrogen. It was further noted by Siddiqui and Sharma[2] that, whereas the Hoffmann degradation of isoconessine gave back the tertiary base quantitatively, the dihydriodides of conessine and iso-conessine by simple heating yielded a common hydrocarbon $C_{21}H_{30}$ with two additional double bonds.

In order to investigate the structural characteristics responsible for this anomaly and the influence of isomerization on the N-stability of the N-methyl groups on the one hand, and the carbocyclic nucleus on the other, a detailed study of several related alkaloids and their halogen acid salts has been undertaken, using the methods of ultraviolet absorption spectroscopy. The present communication deals with the results obtained with conimine, conessine, isoconessimine, their dihydrochlorides, dihydrobromides and dihydroiodides.

Ultra-Violet Absorption Measurements on the Bases and their Halogen Acid Salts

Solutions of strengths 0.1% and 0.01% were prepared and their absorbance was measured at a series of wave-lengths in the range 210 mμ to 400 mμ. Graphs were then drawn between log-absorbance (reduced to C=1.0%) and wave-length in mμ. In the case of the pure bases, *i.e.*, conimine and conessine, the solvent used was re-distilled ethanol, while the halogen acid salts were all soluble in distilled water. From the established structural formulae of the three bases (shown below) which contain only one double bond, no strong diene absorption due to two conjugated double bonds (at 230±5 mμ) is to be expected.

It is seen from the graph drawn for conessine that there is only one broad peak and that too of very low strength at 280±5 mμ (Fig. 1). In case of conimine, two such peaks are observed, also of small height, at 235 mμ and 280 mμ (Fig. 1). When the graphs for the dihydrochlorides of conimine, conessine, and isoconessimine were studied, no important change in the character of the absorption was noticed, except that a low peak at about 240 mμ appears in conessine also, as can be seen from the graphs in Fig. 2. The behaviour of the dihydro-bromide of conessine is similar, with the difference that the small peak, which made its appearance in the dihydrochloride, becomes stronger and sharper (Fig. 3). Its position, 240±5 mμ, is close to the wavelength for the diene absorption.

In contrast with these rather small changes, the graphs for the dihydroiodides of conessine, conimine and isoconessimine all show a remarkable feature, namely that a peak of unexpectedly large height is observed at 225 mμ as shown in Fig. 4.

By comparison with the peak molar absorbance for a diene, *i.e.*, a pair of fully conjugated double bonds, we can calculate the equivalent strength

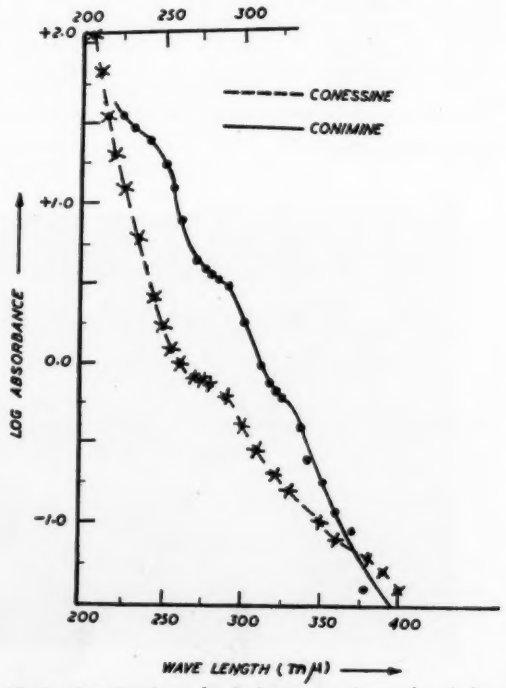


Fig 1. Log absorbance for the bases, conessine and conimine (in ethanol) reduced to a concentration 1%.

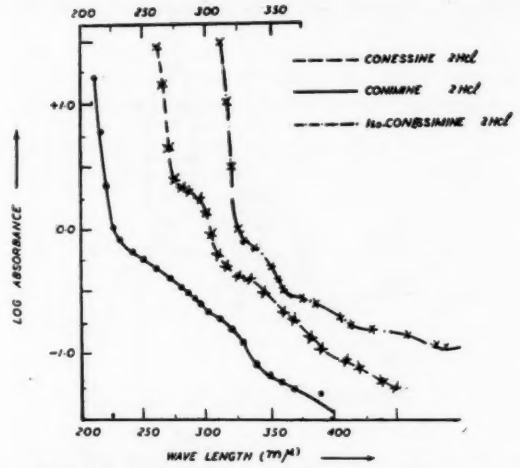


Fig. 2.—Log absorance for dihydrochlorides of the three bases, conessine, conimine and isoconnessimine (in conductivity water) reduced to a concentration of 1% and displaceed successively by 50 mμ.

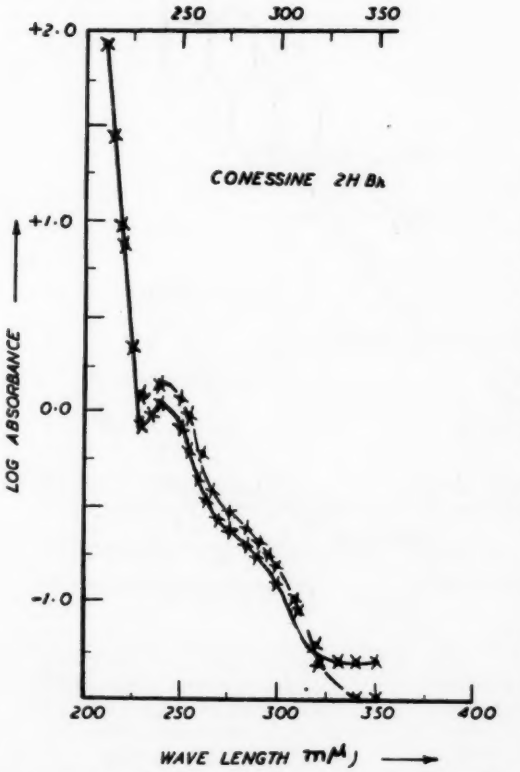


Fig. 3.—Log absorbance for the conessine dihydrobromide (two different samples) reduced to a concentration of 1%.

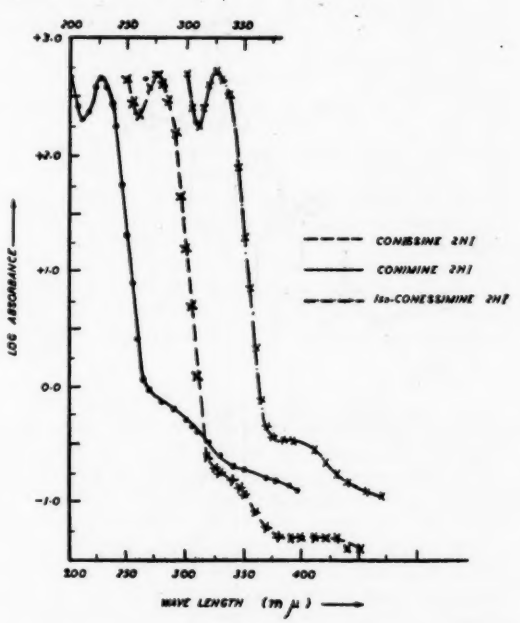
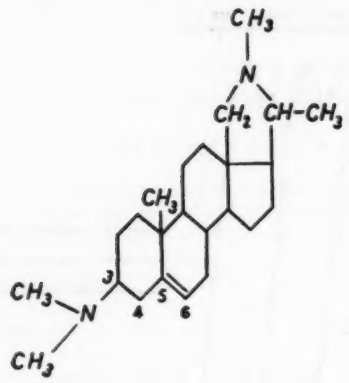


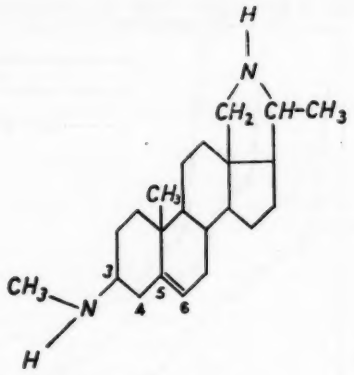
Fig. 4.—Log absorbance for the dihydroiodides of the three bases (in water) reduced to a concentration of 100%.

or degree of conjugation of the bonds corresponding to the observed peak heights in the region of 230 mμ (Table 1). These equivalent strengths for the bases as well as their hydrochlorides and dihydrobromides work out to less than 0.01, which is quite reasonable, because it is usual for several of the bonds to have about 10% double bond character. However, for the dihydroiodide of these three bases *i.e.*, conimine, conessine and isoconessimine, this equivalent strength comes out to be 0.96 ± 0.05, which would require the presence of two double bonds in a position to conjugate fully with each other. Since only one double bond (in the 5,6 position) is known to exist in the carbocyclic nucleus, and the creation of another double bond through the electronic influence of the H I group and the tertiary nitrogen atom is difficult to believe, it was thought probable that the large peak at 225 mμ is not due to the absorption by a diene. In order to clear up this point, it was considered desirable to study the ultra-violet absorption of hydroiodic acid as well as the other halogen acids.*

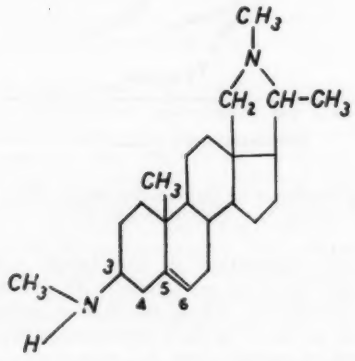
*The International Critical Tables do not give any data for HI while for HBr and HCl, they give only the wavelengths of the absorption maxima.



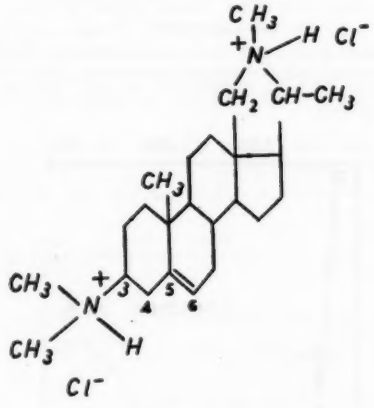
Conessine ($C_{24}H_{40}N_2$)



Conimine ($C_{22}H_{36}N_2$)



Isoconessimine ($C_{23}H_{38}N_2$)



Conessine 2HCl

TABLE I.—ESTIMATED STRENGTH OF PEAKS NEAR 230 mμ.

Conessine	*Conimine*	*Isoconessimine*
No detectable peak near 230 mμ	Peak at 240±5 mμ M.A. ~ 0.3×10^3 units Equiv. strength ~ 0.01	———
Conessine dihydrochloride	*Conimine dihydrochloride*	*Isoconessimine dihydrochloride*
Broad peak at 245±5 mμ M.A. < 0.1×10^3 units Equiv. strength < 0.003 unit	No detectable peak near 230 mμ	Broad peak at 245±5 mμ M.A. < 0.1×10^3 units Equiv. strength < 0.003
Conessine dihydrobromide	*Conimine dihydrobromide*	*Isoconessimine dihydrobromide*
Sharp peak at 240±5 mμ M.A.=0.2×10^3 units Equiv. strength = 0.006.	———	———
Conessine dihydroiodide	*Conimine dihydroiodide*	*Isoconessimine dihydroiodide*
Sharp peak at 225±5 mμ M.A. = 32.4×10^3 units Equiv. strength = 1.02	Sharp peak at 225±5 mμ M.A. = 27.5×10^3 units Equiv. strength = 0.90	Sharp peak at 225±5 mμ M.A. = 29.6×10^3 units Equiv. strength = 0.95.

Note: Tabulated position of diene, triene. and tetraene peaks are 230 ± 5mμ, 270 ± 5 mμ, and 320 ± 10 mμ, respectively.

U.V. Measurements with the Halogen Acids

For comparison with the foregoing observations, the ultra-violet measurements on the halogen acids were carried out on solutions of concentrations 0.002 molar and 0.000,2 molar, which are approximately the same as those used for the free bases and their acid salts. The molar absorbance curves for HCl, HBr, and H I, are all plotted in Fig. 5, from which it can be seen that the absorption peaks for HCl and HBr are all small. Hydroiodic acid, however, in addition to two small peaks, shows a very strong absorption peak at 225 mμ, whose molar absorbance is seen to be nearly 20×10^3, which is about 60% that for the dihydroiodides of conessine, conimine, and isoconessimine. It may thus be concluded that the major part of the anomalously high peak at 225 mμ observed in the dihydroides of these three bases is to be attributed directly to the HI molecular group. It follows that the effect should be similar on both the nitrogen atoms, in agreement with the experimental observation that both nitrogens are eliminated on heating the dihydroiodide.

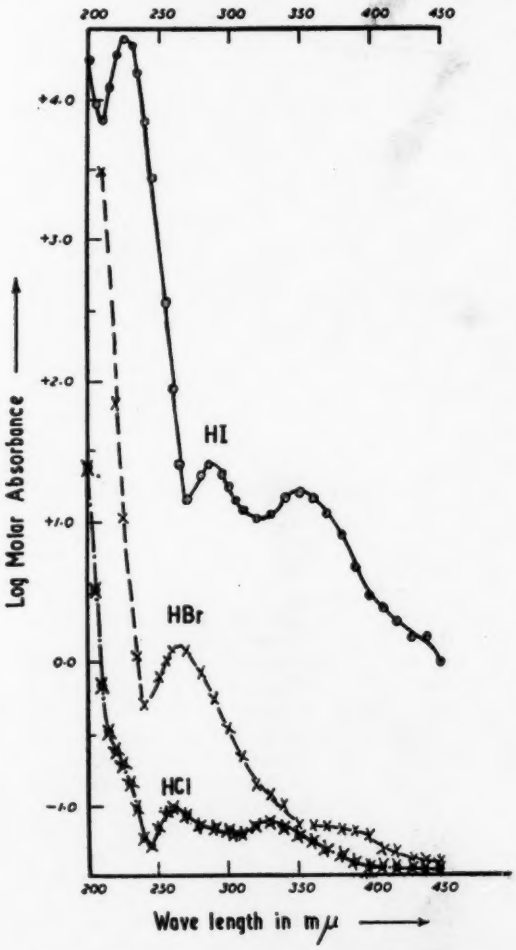


Fig. 5.—Log molar absorbance for the halogen acids, HI, HBr, and HCl (0.002 and 0.000,2 molar solutions in water).

The strong absorption peak would of course mask any small diene peak, occurring at 230±5 mμ, and it is therefore not possible to assert definitely the absence of such a diene absorption. The existence of some such absorption may account for the observed shift of the corresponding peak from 265 mμ in HBr to 240 mμ in conessine dihydrobromide. There is thus a possibility that, due to the formation of the ammonium complex the nitrogen atom may induce a partial double-bond character on the 3,4 bond, which would then conjugate with the known double bond in the 5,6 position. Further investigations, necessary for the elucidation of this point, are in progress, and their results will be reported later.

Acknowledgements

The authors are grateful to Dr. Salimuzzaman Siddiqui for his interest in the problem, and to Dr. Mazhar M. Qurashi for help in planning and executing the ultra violet absorption measurements. Our thanks are also due to Mr. Moqaddas Ali Khan for helping with the preparation of some of the compounds.

References

1. J.v. Braun and R. Schwarz, Ber. **35**, 1279 (1902); J.v. Braun, Ber. **49**, 977, (1916); J.v. Braun, M. Kuhn and S. Siddiqui, Ber., **59**, 1081 (1926).
2. S. Siddiqui, R.H. Siddiqui and V. Sharma, Proc. Indian Acad. Sci. **4A**, 283, (1936).
3. S. Siddiqui and V. Sharma, Proc. Indian Acad. Sci., **6A**, 191-4 (1937).

AN INVESTIGATION OF THE CORRECTION FOR PERIPHERAL HEAT-LOSS FROM THICK SAMPLES IN LEES AND CHORLTON'S DISC METHOD OF DETERMINING THERMAL CONDUCTIVITY

A.K.M. Ahsanullah and Mazhar Mahmood Qurashi

Central Laboratories, Pakistan Council of Scientific and Industrial Research, Karachi

1. Introduction

There are several well-known methods for the determination of the thermal conductivity of poorly conducting building materials like wall-board, light-weight cement, bagasse-board, etc., whose conductivities are in the range of 1×10^{-3} to 0.1×10^{-3} C.G.S. units (cal sec^{-1} $cm^{-1}/^{o}C.$). Of the various methods, Lees and Chorlton's disc method[1] is perhaps the simplest and easiest to set up and manipulate. The method essentially consists in placing a thin disc of radius 'R' of the sample between a freely-suspended brass disc and a steam chest of the same diameter as the sample (Fig. 1), and then measuring the equilibrium temperature θ_b attained by the brass disc.

The thermal conductivity K of the sample is obtained by assuming that the peripheral surface is small and a uniform temperature gradient exists across the thickness of the sample disc (cf. Fig. 2a). On this basis, the heat flowing per unit time across the faces of the sample disc of thickness 'd' is

$$H = K_{expt} \frac{\theta_s - \theta_b}{d} \times \pi R^2 \qquad (1a)$$

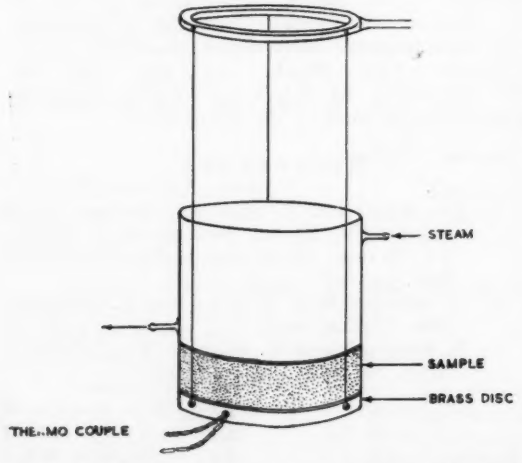


Fig. 1.—Sketch of the experimental arrangement in Lees and Chorlton's disc method for measurement of thermal conductivity of poor conductors.

and this is equated to the heat lost by the brass disc to the surrounding atmosphere (at temperature θ_a), which can be put equal to

$$E_b \times (\pi R^2 + 2\pi R\, d_b) \times (\theta_b - \theta_a) \qquad (1b)$$

where d_b is thickness of the brass disc, and E_b is a constant according to Newton's law of cooling. However, the application of equations (1) is handicapped by the fact that most of the above-mentioned building materials are only available in fair thicknesses, of the order of 0.5 cm. to 2 cm., and therefore a very considerable loss of heat occurs from the peripheral exposed surface of the specimen. For a sample disc 10 cm. in diameter and 1 cm. thick, this peripheral surface has an area of over 30 sq. cm., which is almost one half the area of the surface across which the conduction of heat occurs. It is therefore of great importance to derive a correction for this peripheral loss.

2. The Basic Pattern of Heat-Flow

The calculation for this effect can be made fairly simply, provided the conditions are such that the peripheral loss is smaller than the heat flowing across the sample face in contact with the brass disc. When this condition is satisfied, we can consider the phenomenon as being essentially one of rectilinear conduction with a small superposed distortion of the lines of heat flow caused by the peripheral heat loss. The nature of this distortion is readily found if we remember that unit area on the periphery of the sample disc loses heat to the surroundings at the rate of $E(\theta - \theta_a)$, where θ is the temperature of the area considered and E is a constant according to Newton's law of cooling. This quantity of heat must naturally flow outwards by conduction from just inside the periphery. The actual heat flow near the periphery is thus the resultant of this outward flow and the normal conduction flow parallel to the axis, whence it follows that the actual lines of flow will be inclined outwards from the axis. Also, at points far from the periphery of the sample disc, the lines of flow will be practically axial, so that the inclination of the lines of flow is a function of the distance from the axis of the disc.

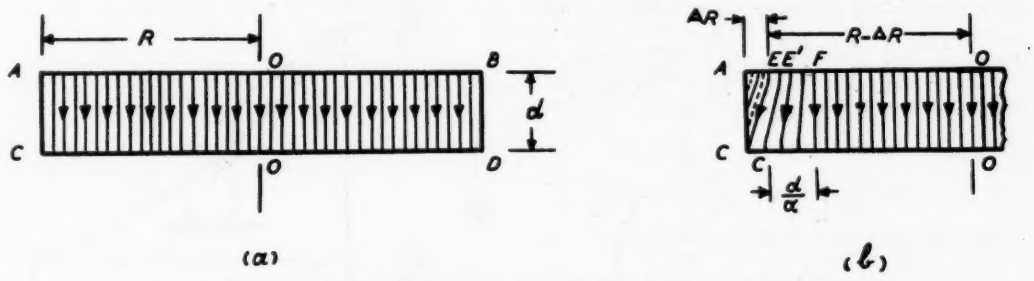


Fig. 2.—Lines of heat flow through the sample disc, assuming (a) ideal case of no peripheral loss, and (b) significant peripheral loss.

It is shown in Appendix I that this inclination is appreciable only within a distance $d/\sqrt{2}$ from the periphery of a disc of thickness 'd'. The ideal and the actual lines of flow are shown schematically in Figs. 2a and 2b, wherein all the lines of flow are parallel to the axis in the immediate neighbourhood of either face of the sample in consequence of the temperature uniformity imposed by the brass disc and the steam-chest. It is seen that only the heat flowing into a circle of radius $(R-\Delta R)$ flows out at the bottom of the sample, while the heat flowing into the annulus between radii R and $R-\Delta R$ ultimately flows out at the periphery of the sample. This feature enables us to calculate the corrections to be made for the peripheral loss. The correction consists of two components : (a) the peripheral loss as such, and (b) the discrepancy caused by the increased length of the tubes of flow due to their inclination to the axis.

3. Derivation of the Correction Formula

Figure 3a shows an enlarged view of a typical tube of flow, and, if we neglect the small curvature near either face of the sample, this tube can be replaced to a sufficient accuracy by the inclined straight lines of Fig. 3b, in which the undistorted axial flow is shown by the vertical broken lines. If the area intercepted by the inclined tube on the median plane XY is δA, then the mean cross-sectional area of this tube is $\delta A \times \cos\phi$ and its length is $d \sec\phi$, where ϕ is the inclination. Therefore the quantity of heat, δH, flowing per unit time through this tube is given by

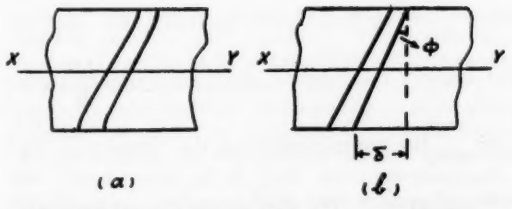


Fig. 3.—Enlarged views of inclined tubes of heat flow (a) exact shape, and (b) approximation suitable for calculations.

$$\delta H = K(\theta_s-\theta_b)\,\frac{\delta A\cos\phi}{d\sec\phi} = \frac{K}{d}(\theta_s-\theta_b)\times\delta A\cos^2\phi$$

$$= \frac{K}{d}(\theta_s-\theta_b)\times\delta A/(1+\delta^2/d^2), \qquad (2)$$

where θ_s is the temperature of steam, θ_b is the temperature of brass disc, K is the thermal conductivity of the sample, and $\delta = d\tan\phi$ is the displacement of one end of the tube of flow relative to the other. It follows that the total heat H flowing out at the bottom of the sample is

$$H = \int \delta H = \frac{K}{d}(\theta_s-\theta_b)\int \delta A/(1+\delta^2/d^2)$$

$$= \frac{K}{d}(\theta_s-\theta_b)\int_0^{R-\frac{1}{2}\Delta R} 2\pi r\,dr/(1+\delta^2/d^2), \qquad (3)$$

where the upper limit of integration is determined by the fact that only the heat flowing upto the inclined line CE in Fig. 2b emerges at the lower face of the sample. Now it is shown in Appendix I that δ/d is zero for $r < \left(R-\frac{\Delta R}{2}-\frac{d}{\sqrt{2}}\right)$, and for larger r is proportional to $\left[\,r-\left(R-\frac{\Delta R}{2}-\frac{d}{\sqrt{2}}\right)\right]$ with a maximum value of $\frac{\Delta R}{d}$ at $r = R-\frac{\Delta R}{2}$ (cf. Fig. 2b). Thus if we put $r_0 = \left(R-\frac{\Delta R}{2}-\frac{d}{\sqrt{2}}\right)$, then we have for $r > r_0$,

$$\delta/d = \frac{r-r_0}{d/\sqrt{2}}\times\frac{\Delta R}{d},$$

and

$$\frac{1}{1+\delta^2/d^2} = 1-\delta^2/d^2+\delta^4/d^4\ldots\ldots\ldots\ldots$$

$$= 1-2\left(\frac{r-r_0}{d}\right)^2\left(\frac{\Delta R}{d}\right)^2+4\left(\frac{r-r_0}{d}\right)^4\left(\frac{\Delta R}{d}\right)^4$$

$$\ldots\ldots\ldots\ldots\ldots\ldots$$

Substitution of this expression into equation (3) gives to sufficient accuracy

$$H = K\,\frac{\theta_s-\theta_b}{d}\left[\int_0^{R-\frac{1}{2}\Delta R} 2\pi r\,dr - \int_0^{R-\frac{1}{2}\Delta R} 4\pi r\left(\frac{r-r_0}{d}\right)^2 \times \left(\frac{\Delta R}{d}\right)^2 dr\right]$$

$$= K\,(\theta_s-\theta_b)\,\frac{\pi R^2}{d}\left[\left(1-\frac{\Delta R}{2R}\right)^2 - \left(\frac{\Delta R}{R}\right)^2 \times \left(\frac{\sqrt{2}}{3}\,\frac{r_0}{d} + \frac{1}{4}\right)\right]$$

$$\simeq K\,(\theta_s-\theta_b)\,\frac{\pi R^2}{d}\left(1-\frac{\Delta R}{2R}\right)\left(1-\frac{\Delta R}{2R} - \frac{\Delta R}{R}\times\frac{\Delta R}{d}\times\frac{\sqrt{2}}{3}\right) \qquad (4)$$

It remains now to obtain an estimate of the quantity ΔR. We notice from Fig. 2b that all the flow lines to the left of the line EC terminate on the periphery of the sample disc, which means that all the heat flowing in at the annulus AE of the disc is to be equated to the heat lost from the periphery, which latter is given by

$$2\pi R \times d \times E \times (\bar{\theta}-\theta_a), \qquad (5)$$

where θ_a is the ambient temperature and $\bar{\theta}$ is the mean temperature of the peripheral surface. This mean temperature will be somewhat less than $\frac{1}{2}(\theta_s+\theta_b)$ because the temperature distribution is not truly linear, but is changed by the surface loss into the form shown in Fig. 4. Assuming a linear variation of $\partial\theta/\partial x$ (which is close to the truth), we may put

$$\frac{\partial\theta}{\partial x} = \frac{\overline{\partial\theta}}{\partial x} + \frac{\partial^2\theta}{\partial x^2}\left(x-\frac{d}{2}\right) = \frac{\theta_b-\theta_s}{d} + \frac{\partial^2\theta}{\partial x^2}\left(x-\frac{d}{2}\right),$$

which integrates to

$$\theta = \theta_s + \frac{\theta_b-\theta_s}{d}x + \frac{1}{2}\,\frac{\partial^2\theta}{\partial x^2}\,x\,(x-d) \qquad (6a)$$

and gives finally

$$\bar{\theta} = \frac{1}{d}\int_0^d \theta\,dx = \tfrac{1}{2}(\theta_s+\theta_b) - \frac{d^2}{12}\,\frac{\partial^2\theta}{\partial x^2} \qquad (6b)$$

Thus the heat loss from the peripheral surface of the sample becomes

$$2\pi R \times d \times E\left(\tfrac{1}{2}(\theta_s+\theta_b) - \theta_a - \frac{d^2}{12}\,\frac{\partial^2\theta}{\partial x^2}\right), \qquad (7)$$

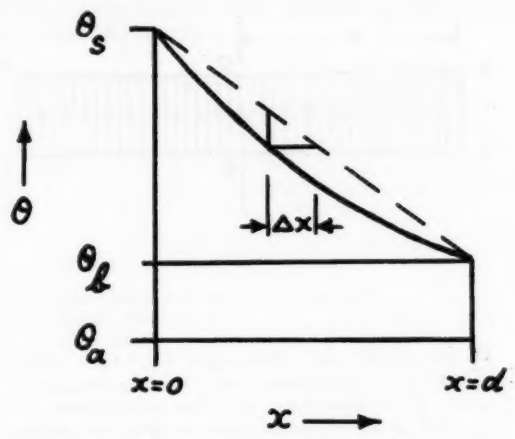


Fig. 4.—Typical temperature distribution on the peripheral surface of the sample.

while the heat flowing into the annulus AE is to a good approximation given by

$$K\pi\left(R^2-(R-\Delta R)^2\right)\left(-\frac{\partial\theta}{\partial x}\right)_{x=0}$$

$$= 2\pi R K \times \Delta R\left(1-\frac{\Delta R}{2R}\right)\left(\frac{\theta_s-\theta_b}{\sqrt{d^2+(\Delta R)^2}} + \frac{d}{2}\,\frac{\overline{\partial^2\theta}}{\partial x^2}\right)$$

$$= 2\pi R\,\frac{K}{d}\,\Delta R\left(1-\frac{\Delta R}{2R}\right)\left(\frac{\theta_s-\theta_b}{\sqrt{1+(\Delta R/d)^2}} + \frac{d^2}{2}\,\frac{\partial^2\theta}{\partial x^2}\right) \qquad (8)$$

On equating the two expressions (7) and (8), and transposing, we get for the maximum inclination of the flow lines

$$\phi_{max} = \frac{\Delta R}{d} = d\,\frac{E}{K} \times \frac{\frac{1}{2}(\theta_s+\theta_b)-\theta_a-\frac{d^2}{12}\,\partial^2\theta/\partial x^2}{(\theta_s-\theta_b)\left(1-\frac{1}{2}(\Delta R)^2/d^2\right)+\frac{1}{2}\partial^2\theta/\partial x^2} \Big/ \left(1-\frac{\Delta R}{2R}\right) \qquad (9)$$

Since $\frac{\Delta R}{d}$ is itself a small quantity, we can at first neglect the equally small quantities $\frac{\Delta R}{R}$ and $\left(d^2\,\frac{\partial^2\theta}{\partial x^2}\right)$ in the expression on the right-hand side of equation (9). We thus get to a good approximation,

$$\phi_{max} = \frac{\Delta R}{d} = d\,\frac{E}{K}\,[\tfrac{1}{2}\,(\theta_s+\theta_b)-\theta_x]/(\theta_s-\theta_b)$$

$$=d\,\frac{E}{K}\left(\frac{1}{2}+\frac{\theta_b-\theta_x}{\theta_s-\theta_b}\right) \qquad (10)$$

Simplification of equation (4) and substitution of the above expression for ΔR now gives to a sufficient accuracy

$$H = K(\theta_s-\theta_b)\,\frac{\pi R^2}{d}$$

$$\times\left[1-\frac{\Delta R}{R}\left(1+\frac{\Delta R}{d}\left(\frac{\sqrt{2}}{3}-\frac{d}{4R}\right)\right)\right]$$

$$=(\theta_s-\theta_b)\,\frac{\pi R^2}{d}\left[K-\frac{E}{R}\,d^2\left(\frac{1}{2}+\frac{\theta_b-\theta_a}{\theta_s-\theta_b}\right)\right.$$

$$\left.\times\left(1+\frac{E}{2K}\,d\left(\frac{1}{2}+\frac{\theta_b-\theta_a}{\theta_s-\theta_b}\right)\left(\frac{\sqrt{2}}{1.5}-\frac{d}{2R}\right)\right)\right]$$

and finally, $K_{expt} = \dfrac{H}{\theta_s-\theta_b}\times\dfrac{d}{\pi R^2}$

$$= K-\frac{E}{R}\,d^2_{reduced}, \qquad (11a)$$

with $d^2_{reduced} = \left(1+\dfrac{E}{2K}d\left(\dfrac{1}{2}+\dfrac{\theta_b-\theta_a}{\theta_s-\theta_b}\right)\right.$

$$\left.\times\left(\frac{\sqrt{2}}{1\cdot5}-\frac{d}{2R}\right)\right)\left(\frac{1}{2}+\frac{\theta_b-\theta_a}{\theta_s-\theta_b}\right)^{d^2}$$

$$=(1+\mu)\left(\frac{1}{2}+\frac{\theta_b-\theta_a}{\theta_s-\theta_b}\right)d^2 \qquad (11b)$$

where μ is nearly constant and less than 0.1, cf. Table 2. If the neglected quantities, $\dfrac{\Delta R}{R}$ and $d^2\,\dfrac{\partial^2\theta}{\partial x^2}$, are also taken into account, then we find in Appendix II that the factor $(1+\mu)$ is to be replaced by $(1-\mu')$, where μ' is again nearly constant and $\sim$ 0.1. μ(or μ') and therefore $d^2_{reduced}$ can be easily calculated provided we have an approximate value of E, which can be got from two experimental determinations of K with different values of the thickness d. The formula (11) then gives us the desired relation between the actual value of K and the experimentally determined value. The correction to the experimental value of K is seen to be inversely proportional to the radius R of the sample and approximately proportional to the square of its thickness, d. This is as it should be, because the dependence on d is two-fold, the peripheral loss increases linearly with d, while the heat conduction across the disc is inversely proportional to d. The influence of the factors $\left(\dfrac{1}{2}+\dfrac{\theta_b-\theta_a}{\theta_s-\theta_b}\right)$, however, needs some elucidation.

4. Behaviour of the Correction Formula

Let us here examine the formula (1a) used for calculating the value of K_{expt} from the equilibrium temperature, θ_b, of the brass disc, wherein it is assumed that there is no peripheral heat loss from the sample. Equations (1) give us

$$K_{expt} = E_b\,\frac{\pi R^2+2\pi R d_b}{\pi R^2}\times d\,\frac{\theta_b-\theta_a}{\theta_s-\theta_b}$$

$$= E'_b\times d\,\frac{\theta_b-\theta_a}{\theta_s-\theta_b} \qquad (12)$$

where $E'_b = E_b\,(1+2d_b/R)$.
It follows that

$$\left(\frac{1}{2}+\frac{\theta_b-\theta_a}{\theta_s-\theta_b}\right)=\frac{1}{2}+\frac{K_{expt}}{dE'_b}.$$

Substitution of this expression into equation (11b) gives

$$d^2_{reduced} = \left(1+\left(\frac{\sqrt{2}}{1.5}-\frac{d}{2R}\right)\right.$$

$$\left.\times\left(\frac{1}{2}+\frac{K_{expt}}{d\,E'_b}\right)\frac{Ed}{2K}\right)\left(\frac{1}{2}+\frac{K_{expt}}{dE'_b}\right)\times d^2$$

$$=(1+\mu)\left(\frac{d}{2}\times\frac{K_{expt}}{E'_b}\right)\times d \qquad (13a)$$

where $\mu=\left(\dfrac{\sqrt{2}}{1.5}-\dfrac{d}{2R}\right)\left(\dfrac{d}{2}+\dfrac{K_{expt}}{E'_b}\right)\times\dfrac{E}{2K}$

$$\simeq \frac{1}{3}\,\frac{E}{E'_b} \qquad (13b)$$

Finally we get from equation (11a),

$$K_{expt}=K-\frac{E(1+\mu)}{R}\,d\left(\frac{d}{2}+\frac{K_{expt}}{E'_b}\right) \qquad (14)$$

Inclusion of the higher order terms neglected in equation (10) will as before result in the replacement of μ by $-\mu'$, where μ and μ' are both $\sim$0.1.

For thin samples, *i.e.*, when d is small, equation (14) reduces to

$$K_{expt} = K - \frac{E}{R}\, d\, \frac{K_{expt}}{E'_b}\left(1 + \frac{1}{3}\frac{E}{E'_b}\right)$$

$$\simeq K\left(1 - \frac{d}{R} \times \frac{E}{E'_b}\right) \qquad (15)$$

which shows that with the experimental arrangement of Fig. 1, the values of K determined for thin samples are too low by a factor of $\left(1 - \frac{d}{R} \times \frac{E}{E'_b}\right)$.

This is a direct consequence of the fact that even for the thinnest samples, the maximum inclination of the lines of heat-flow does not become zero but rather tends to a constant value. From equations (10) and (12), we have for the inclination at the periphery

$$\left(\phi_{max}\right)_{d\to 0} = \left(\frac{\Delta R}{d}\right)_{d\to 0}$$

$$= \frac{E}{K}\left[\, d\left(\frac{1}{2} + \frac{\theta_b - \theta_a}{\theta_s - \theta_b}\right)\right]_{d\to 0}$$

$$= \frac{E}{K}\left(\frac{d}{2} + \frac{K_{expt}}{E'_b}\right)_{d\to 0} \simeq \frac{E}{E'_b},$$

which is small in the practical examples discussed below, but is never zero.

In order to study the variation of K_{expt} with the thickness d, we take the following typical values for the constants in equation (14) :

$K = 0.4 \times 10^{-3}$ (cal sec^{-1} cm^{-1}/°C), $E = 0.1 \times 10^{-3}$, $E'_b = 0.3 \times 10^{-3}$, $R = 5$ cm.,

and if we take $K_{expt} \simeq 0.3$, our relation becomes approximately

$$1000\, K_{expt} = 0.4 - \frac{0.1}{5}\, d\left(\frac{d}{2} + \frac{0.3}{0.3}\right)$$

$$\times\left(1 + \left(0.943 - \frac{d}{2\times 5}\right)\left(\frac{1}{2} + \frac{0.3}{0.3d}\right) \times \frac{0.1}{2\times 0.4} d\right)$$

$$= 0.4 - \frac{d}{50}\left(1 + \frac{d}{2}\right)\left(1 + \left(0.943 - \frac{d}{10}\right)\right.$$

$$\left.\times\left(0.125 + \frac{d}{16}\right)\right).$$

The values got from this relation are shown in Table 1 below together with the corresponding values of d and d^2. For small d, the change in K is roughly proportional to d, while for large d, the variation depends more nearly on d^2. If a simple working formula is sought, it is probably accurate enough to try the compromise

$$K_{expt} = K - a\, d^{\frac{3}{2}} \qquad (16)$$

where 'a' will be of the order of

$$\frac{E}{R}\left(\frac{K}{E'_b} \times \frac{R \times E}{E'_b}\right)^{\frac{1}{2}} \sim \frac{E}{R}.$$

Such a treatment of the calculated data of Table 1 is shown in Fig. 5 in which the values for d = 0 and 0.5 cm. are taken as unknown. The extrapolation of the straight line to $d^{\frac{3}{2}} = 0$ gives $K = 0.406 \times 10^{-3}$ (cal sec^{-1} cm.$^{-1}$/°C.), which is within 1½ % of the correct value, so that this simplified equation (16) should be acceptable within the usual limits of experimental error for K, *i.e.*, about 0.01×10^{-3} or a little more.

5. Experimental Verification

For experimental work, either this above simplified technique or the more elaborate equation (11) can be used. In order to estimate the relative merits of the two procedures, an analysis is presented for the results of a series of experiments performed in this laboratory with "Celeron" discs of different thicknesses from half an inch upto almost two inches. Several discs 10 cm. in diameter were cut and accurately machined out of a quarter-inch thick sheet of "Celeron," which is a variety of hard laminated insulating material. Composite discs of various thicknesses were made up out of these by putting together 2, 3, 4, etc., of these ¼" thick discs. The thermal conductivity of these composite discs was determined with the apparatus of Fig. 1 by measuring the equilibrium temperature of the brass disc in each case. The mean results for two sets of determinations are shown in Table 2 along with other relevant data.

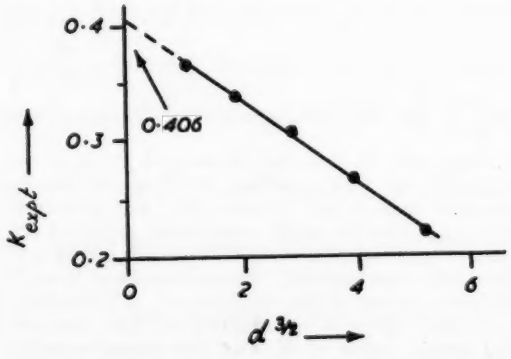


Fig. 5.—Plot of typical theoretical values of K_{expt} against $d^{3/2}$ to show approximate linearity.

TABLE 1.—CALCULATED VARIATION OF K_{EXPT} IN A TYPICAL CASE.

d (cm.) : ..	0	0.5	1.0	1.5	2.0	2.5	3.0
$1000 \times K_{expt}$ ..	0.400	0.38_6	0.36_5	0.33_8	0.30_5	026_5	0.22_0
d^2(cm.^{2}) ..	0	0.25	1.00	2.25	4.00	6.25	9.00

TABLE 2.—EXPERIMENTAL DATA FOR "CELERON" DISCS UPTO 2 INCHES THICK.

"Celeron" Discs : R = 5.0 cm. E'_b for brass disc = 0.290×10^{-3}

No. of discs	d (cm.)	θ_b (°c.)	θ_a (°c.)	$\frac{1}{2}+\frac{\theta_b-\theta_a}{\theta_s-\theta_b}$	$\frac{d}{2}+d\frac{\theta_b-\theta_a}{\theta_s-\theta_b}$	K_{expt} (cal $sec^{-1}cm^{-1}/°c.$)	d^2 (cm.^{2})	$1+\mu$	$d^2_{reduced}$	$d^{3/2}$
2	1.27	65.7	29.8	1.55	1.97	$0.40_6 \pm 01_5$	1.61	1.064	2.7	1.4_3
3	1.90	61.2	30.0	1.31	2.49	$0.39_6 \pm 00_8$	3.60	1.07_5	5.1	2.6_2
4	2.54	54.0	29.0	1.05	2.67	$0.38_4 \pm .008$	6.44	1.07_4	7.3	4.0_5
5	3.18	49.4	29.3	0.90	2.86	$0.34_7 \pm .007$	10.1_0	1.07_1	9.7	5.6_6
6	3.81	44.6	28.2	0.80	3.05	$0.31_3 \pm .001$	14.5	1.06_9	12.4	7.4_5
7	4.44	41.9	28.4	0.74	3.28	$0.290 \pm .006$	19.7	1.06_6	15.5	9.3_5

Note :—Experimental values for one disc have been omitted because with our apparatus, the temperature, θ_b, becomes so high that the Newton's law of cooling is no longer accurately applicable to the heat lost by the brass disc.

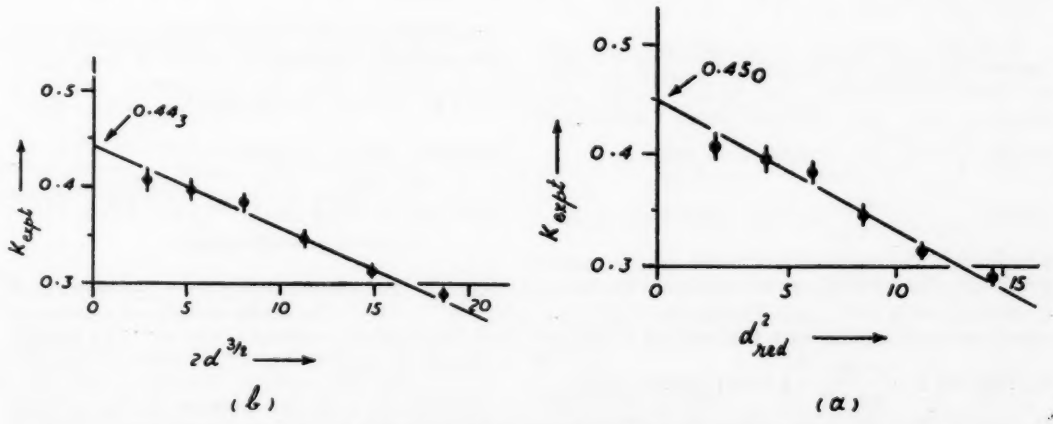


Figure 6. Experimental graphs for "Celeron" discs upto 5 cm. thickness. The measured values (K_{expt}) of theramal conductivity are plotted against (a) d^2reduced, and (b) $d^{3/2}$, and thus extrapolated to zero sample thickness.

First we try to fit to the experimental data, the more accurate equations (11), according to which the graph for K_{expt} against $d^2_{reduced}$ should be a straight line. In order to calculate $d^2_{reduced}$, we require an approximate value of E (cf. equation 11(b)). This can be done by assuming $d^2_{reduced} = d^2$ in the first instance and substituting two of the values from Table 2 into equation 11(a). Thus

$$\left.\begin{aligned} 0.406\times 10^{-3} &= K - \frac{E}{R}\times 1.61 \\ 0.290\times 10^{-3} &= K - \frac{E}{R}\times 19.7 \end{aligned}\right\}$$

whence $\frac{E}{R} = \frac{0.116}{18.1}\times 10^{-3} = 0.0064\times 10^{-3}$,

and therefore $E = 5\times 0.0064\times 10^{-3} = 0.032\times 10^{-3}$

$$\text{and } \frac{E}{2K} = 0.032/0.8 = 0.04.$$

This enables us to calculate the factor $(1+\mu)$ given in column 9 of Table 2 and finally $d^2_{reduced}$, which is given in column 10. The graph for K_{expt} against $d^2_{reduced}$ is shown in Fig. 6a, in which the short vertical lines through the plotted points indicate their estimated standard errors. The straight line drawn in the figure is seen to fit the experimental points very well, the deviations from the straight line being about equal to the standard errors on the average. This means that equation (11) holds within the limits of experimental accuracy. The corrected value of K found by extrapolation to $d^2_{reduced} = 0$ is 0.450×10^{-3}.

A further refinement is to get the accurate value of E/R from the slope of the graph of Fig. 6a, and to recalculate the factor $(1+\mu)$ and $d^2_{reduced}$ with this accurate value. From the graph, $\frac{E}{R} = \frac{0.160}{15}\times 10^{-3}$, *i.e.*, $E = 0.053\times 10^{-3}$,

whence $\frac{E}{2K} = \frac{0.053}{2\times 0.45} = 0.059$, which is to be compared with the approximate value of 0.04 used previously. Since $\frac{1}{2}E/K$ enters only in the quantity, μ, the values of $d^2_{reduced}$ calculated with this more accurate estimate of $\frac{1}{2}E/K$ will be increased by a factor $\left(1+\frac{0.059}{0.040}\times\mu\right)\Big/(1+\mu)$, where $\mu\sim 0.07$. It follows that the new values are all almost exactly 1.03 times the old ones, so that the graph will be similar to the previous one, but with the abcissa expanded in this ratio. The value of K extrapolated to $d^2_{reduced} = 0$ will therefore be the same as before. Similar remarks apply to the replacement of $(1+\mu)$ by $(1-\mu')$ as a further refinement (Appendix II).

For comparison with the foregoing process, we show a plot of the values of K_{expt} against $d^{\frac{3}{2}}$ in Fig. 6b. The points can again be fitted by a straight line within the limits of experimental error, and the value of K obtained by extrapolation to $d = 0$ is $0.44_3\times 10^{-3}$, which is lower than the corrected value (got from Fig. 6a) by 0.007×10^{-3}, *i.e.*, by about 2%.

6. Examples of Applications and Generalization

We take as an illustration, the evaluation of the correction for peripheral heat loss in a typical case, namely that of a cellular cement disc 10 cm. in diameter and 1.2 cm. thick, the density being 0·35 g./ml. For such a disc, the value obtained experimentally with the apparatus of Fig. 1 was

$$K_1 = (K_{expt})_{d=1.2} = 0{\cdot}11_0\times 10^{-3}\pm 0{\cdot}005\times 10^{-3}$$

cal sec^{-1} $cm^{-1}/°C$.

Two approaches are now possible. Firstly we may proceed as with the "Celeron" discs, that is, the experimental determination may be repeated with two such cement discs placed one above the other (thus making a single disc of twice the thickness). This gave a value of

$$K_2 = (K_{expt})_{d=2.4} = 0.08_7\times 10^{-3}\pm 0{\cdot}005\times 10^{-3}$$

cal sec^{-1} $cm.^{-1}/°C$.

With the help of the already tested equation (11), and using the approximate value of 0.06×10^{-3} for E, together with the experimental values of θ_b and θ_a, we easily get for the correction to K_1,

$$\text{correction} = (K_{correct} - K_1) = \frac{K_1 - K_2}{[(d^2_{reduced})_2/(d^2_{reduced})_1]^{-1}} = \frac{0{\cdot}02_3\pm 0{\cdot}00_7}{(4{\cdot}55/1{\cdot}67)-1}\times 10^{-3}$$
$$= (0{\cdot}013\pm 0{\cdot}004)\times 10^{-3},$$

whence K corrected $= 0.12_3\times 10^{-3} \pm 0.007\times 10^{-3}$ cal sec^{-1} $cm.^{-1}/°C$, which is in good agreement with the tabulated value of 0.14×10^{-3} for cement of the given specification.

Secondly we note that because the loss of heat from the periphery of the sample is mostly due to convection rather than radiation as such, the value of the constant E in equation (5) will be in-

dependent of the nature of the sample. Therefore we can use the experimental value (got from the data on "Celeron" discs) to calculate the correction for any sample with the help of equations (11) or (14). Substitution of the values for E, E_b, and R into equation (14) gives for our apparatus,

$$K_{expt} = K - \frac{0.053 \times 1.1}{5000}\left(\frac{d}{2}+\frac{K_{expt}}{0.29}\right)d,$$

$$= K - \frac{0.0116}{1000}\left(\frac{d}{2}+\frac{K_{expt}}{0.29}\right)d, \qquad (17)$$

if we take $(1+\mu)$ as 1.1 which is quite accurate for our work.* The only quantities needing measurement are d and K_{expt} for the sample disc. Substitution of their values into the above equation gives directly

$$K = 0.11_0 \times 10\text{-}3 + 0.0116 \times 10\text{-}3\left(\frac{1.2}{2}+\frac{0.11}{0.29}\right) \times 1.2$$

$$= (0.11_0 + 0.014) \times 10\text{-}3 = 0.124 \times 10\text{-}3$$

cal sec^{-1} cm.$^{-1}$/°C,

which is in excellent agreement with the previous figure of $0.12_3 \times 10\text{-}_3$. This agreement provides experimental verification of the fact that, once the apparatus has been calibrated with uniform samples of various thicknesses, the correction for peripheral loss can be directly calculated for an unknown sample from a *single determination* of the thermal conductivity to an accuracy of about 0.01×10^{-3} or better.

Acknowledgement

The authors are indebted to Mr. Sadrul Hasan Rizvi for help with some of the earlier experiments.

Reference

1. B.L. Worsnap and H.T. Flint, *Advanced Practical Physics for Students* (Methuen & Co. Ltd., London, 1941), sixth edition, p. 227.

Appendix I

The Variation of the Inclination, ϕ, of the Tubes of Flow

The precise value of this inclination will depend on the radial distance, r, from the axis of the sample disc, and will be determined by the boundary conditions, *i.e.*, by the heat loss and the temperature gradient on the peripheral surface. An expression for the dependence of ϕ on r can be obtained by successive approximations. Let us assume at first that ϕ is proportional to r, *i.e.*, $\phi = \frac{r}{R}\phi_{max}$, cf. the straight line OM in Fig. 7.

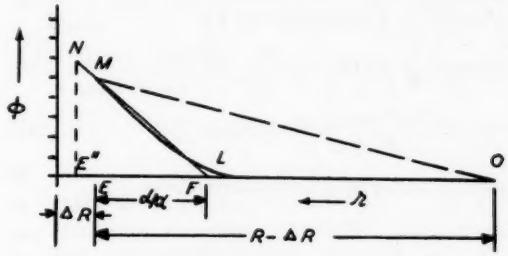


Fig. 7.—Successive approximations to the inclination (ϕ) of the lines of heat flow as a function of the radial distance. First approximation: straight line OM; second approximation: two straight lines, OF and FM; actual: curve OL M.

Since the isothermal surfaces are normal to the lines of flow, the equation of a section made by the plane of the paper through the isothermal surface for $\theta = \frac{1}{2}(\theta_s + \theta_b)$ will be given by (cf. Figs. 3b & 7)

$$-\frac{dx}{dr} = \tan\phi \simeq \frac{r}{R}\phi_{max} \text{ and } (x)_{r=0} = \frac{d}{2}.$$

whence $x = \frac{d}{2} - \frac{1}{2}\frac{r^2}{R}\phi_{max}$,

and $\Delta x = \left(\frac{d}{2} - x\right) = \frac{1}{2}\frac{r^2}{R}\phi_{max}$.

Thus the point on the periphery corresponding to the temperature $\frac{1}{2}(\theta_s + \theta_b)$ is shifted from the middle (*i.e.*, x=d/2) by a distance

$$(\Delta x)_{max} = \frac{1}{2}\frac{R^2}{R}\phi_{max} = \frac{1}{2}R\phi_{max} = \frac{R}{d} \times \frac{\Delta R}{2}, \qquad (18a)$$

because $\phi_{max} = \Delta R/d$, cf. Fig. (2). Another estimate of $(\Delta x)_{max}$ can be got by considering the temperature distribution along the tube of flow ECC′ E′ near the periphery (Fig. 2). For this tube,

$$\frac{\text{Corss-section at CC}'}{\text{Cross-section at EE}'} = \left(\frac{R}{R-\Delta R}\right)^2 \simeq 1+2\frac{\Delta R}{R}.$$

Now, for any tube, the temperature gradient $\partial\theta/\partial s$ is inversely proportional to the cross-section, so that

*It should be noted that when we determine E from the slope of the graph of Fig. 6 *a*, the value obtained includes any factors that may be introduced by further refinements such as those of Appendix II. Thus the replacement of $(1+\mu)$ by $(1-u')$ is automatically taken care of.

$$\frac{(\partial\theta/\partial x)E}{(\partial\theta/\partial x)C} = \frac{\text{Cross-section at CC}'}{\text{Cross-section at EE}'} = 1 + 2\frac{\Delta R}{R},$$

whence $\frac{d}{R}\times 2\Delta R = 2d\frac{\Delta R}{R} = \frac{(\partial\theta/\partial x)_C - (\partial\theta/\partial x)_E}{-\partial\theta/\partial x}$

$$\times d = \frac{d^2\partial^2\theta/\partial x^2}{-\partial\theta/\partial x} = 8\,(\Delta x)_{max}, \qquad (18b)$$

because from Fig. 4 and equation (6a), $\frac{1}{8}d^2\frac{\partial^2\theta}{\partial x^2} = -\frac{\partial\theta}{\partial x}$ $\times (\Delta x)_{max}$. The two estimates for $(\Delta x)_{max}$ can be brought into agreement only if $R = d/\sqrt{2}$, which suggests that the influence of the peripheral loss does not extend all the way inside the disc but is appreciable only to a distance of the order of $d/\sqrt{2}$ inwards from the periphery. This means that the inclination, ϕ, will vary somewhat as shown by the curve OLM in Fig. 7, instead of the straight line OM as initially assumed. For the purposes of calculating the integral of equation (3), a fairly good approximation is got by replacing this curve by the two straight lines OF ($\phi = 0$) and FM, where the distance $EF = d/\alpha$ and α is to be determined from the boundary conditions. Thus the maximum displacement of the isothermal for $\theta = \frac{1}{2}(\theta_s + \theta_b)$ is

$$(\Delta x)_{max} = \frac{d}{2\alpha}\,\phi_{max} = \frac{d}{2\alpha}\times\frac{\Delta R}{d} = \frac{\Delta R}{2\alpha} \qquad (19)$$

For the flow along the tube of flow near EC, we have, analogously to equation (18b), the result

$$2\left[\left(\frac{d/\alpha+\Delta R}{d/\alpha}\right) - 1\right] = 2\frac{\Delta R}{d/\alpha} = \frac{8}{d}(\Delta x)_{max},$$

whence $(\Delta x)_{max} = \frac{\alpha}{4}\times\Delta R \qquad (20)$

By equating equations (19) and (20), we get

$$\frac{1}{2\alpha} = \frac{\alpha}{4}, \qquad i.e.\ \alpha = \sqrt{2}.$$

Thus the distance $EF = \frac{d}{\alpha} = d/\sqrt{2}. \qquad (21)$

Appendix II

Effect of Quantities Neglected in Equation (10)

In order to obtain the departure from linearity of the peripheral temperature distribution of the sample disc, we must calculate the value of $(\Delta x)_{max}$ on the peripheral surface for the middle isothermal, *i.e.*, for $\theta = \frac{1}{2}(\theta_s + \theta_b)$. This isothermal cuts the peripheral surface at $x \sim \frac{d}{2}$, and the line of flow through this intersection will therefore originate near E″, the middle of AE, Figs. 2, 7, at distance $\frac{\Delta R}{2}$ inwards from the periphery. To get the value of $(\Delta x)_{max}$ for this line of flow, we need the variation of ϕ over the region EE″, of extent $\frac{\Delta R}{2}$.

Since all the heat flowing into the annulus AE is ultimately lost to the surroundings by the peripheral surface, whose temperature rises from bottom to top, it is clear that the inclination of the lines of flow will increase as we move from E towards A. The distance $EE'' = \frac{\Delta R}{2}$ being ordinarily much smaller than d, we may, to a first approximation, assume that over this range ϕ continues to increase according to the line MN, which is an extrapolation of the straight line FM. On this basis, we get, as in equation (19),

$$\left[(\Delta x)_{max}\right]_{periphery} = \frac{1}{2}\left(\frac{d}{\sqrt{2}} + \frac{\Delta R}{2}\right) \times \left[\phi_{max}\times\frac{d/\sqrt{2}+\Delta R/2}{d/\sqrt{2}}\right]$$

$$= \frac{\Delta R}{2\sqrt{2}}\left(1 + \frac{\Delta R}{d\sqrt{2}}\right)^2 \sim \frac{\Delta R}{2\sqrt{2}}\left(1+\sqrt{2}\frac{\Delta R}{d}\right)$$

It follows from equation 6(a) that

$$\left(\frac{d^2}{2}\frac{\partial^2\theta}{\partial x^2}\right)\Big/\left(\theta_s-\theta_b\right) = \frac{4}{d}\left[(\Delta x)_{max}\right]_{periphery}$$

$$= \sqrt{2}\,\frac{\Delta R}{d}\left(1+\sqrt{2}\frac{\Delta R}{d}\right) \qquad (22)$$

Substitution of this result into equation (9) gives us

$$\frac{\Delta R}{d} = \phi_{max} = \frac{E}{K}d \times \frac{\frac{1}{2}(\theta_s+\theta_b)-\theta_o-(\theta_s-\theta_b)\frac{\sqrt{2}}{6}\frac{\Delta R}{d}\left(1+\sqrt{2}\frac{\Delta R}{d}\right)}{(\theta_s-\theta_b)\left(1-\frac{1}{2}\left(\frac{\Delta R}{d}\right)^2+\sqrt{2}\frac{\Delta R}{d}\left(1+\sqrt{2}\frac{\Delta R}{d}\right)\right)} \div \left(1-\frac{\Delta R}{2R}\right)$$

$$= d\,\frac{E}{K}\left(\frac{1}{2} + \frac{\theta_b-\theta_o}{\theta_s-\theta_b} - \frac{\sqrt{2}}{6}\frac{\Delta R}{d}\left(1+\sqrt{2}\frac{\Delta R}{d}\right)\right)$$

$$: \left(1+\frac{3}{2}\left(\frac{\Delta R}{d}\right)^2+\sqrt{2}\frac{\Delta R}{d}-\frac{\Delta R}{2R}\right)$$

$$\sim d\frac{E}{K}\left(\frac{1}{2}+\frac{\theta_b-\theta_o}{\theta_s-\theta_b}\right)\left(\left(1-\frac{\sqrt{2}}{4}\frac{\Delta R}{d}+\frac{1}{2}\left(\frac{\Delta R}{d}\right)^2\right)\right.$$

$$\left.\times\left(1-\sqrt{2}\frac{\Delta R}{d}+\frac{1}{2}\left(\frac{\Delta R}{d}\right)^2+\frac{\Delta R}{2R}\right)\right)$$

$$\sim d\frac{E}{K}\left(\frac{1}{2}+\frac{\theta_b-\theta_o}{\theta_s-\theta_b}\right)\left(1-1.77\frac{\Delta R}{d}\right.$$

$$\left.+\frac{3}{2}\left(\frac{\Delta R}{d}\right)^2+\frac{\Delta R}{2R}\right)$$

$$=d\frac{E}{K}\left(\frac{1}{2}+\frac{\theta_b-\theta_o}{\theta_s-\theta_b}\right)(1-\varepsilon) \qquad (23)$$

Comparison of this expression with equations (10) and (11) then gives after a little algebra

$$d^2_{\text{reduced}}=\left(1+\frac{Ed}{2K}(1-\varepsilon)\left(\frac{1}{2}+\frac{\theta_b-\theta_o}{\theta_s-\theta_b}\right)\left(\frac{\sqrt{2}}{1.5}-\frac{d}{2R}\right)\right)\left(\frac{1}{2}+\frac{\theta_b-\theta_o}{\theta_s-\theta_b}\right)(1-\varepsilon)d^2,$$

which simplifies sufficiently accurately to

$$d^2_{\text{reduced}}=\left(1-\frac{Ed}{2K}\left(\frac{1}{2}+\frac{\theta_b-\theta_o}{\theta_s-\theta_b}\right)\left(2{\cdot}6-\frac{d}{2R}\right)\right)\left(\frac{1}{2}+\frac{\theta_b-\theta_o}{\theta_s-\theta_b}\right)d^2$$

$$=(1-\mu')\left(\frac{1}{2}+\frac{\theta_b-\theta_o}{\theta_s-\theta_b}\right)d^2 \qquad (24)$$

where μ' is approximately independent of d and is of the order of 0.1.

CHEMICAL INVESTIGATION OF NEEM-SEEDS

A.M. Ahsan and George Hahn

Central Laboratories, Pakistan Council of Scientific and Industrial Research, Karachi

In view of the enormous importance attached to the therapeutic properties of the various parts of the Neem-tree (*Azadirachta Indica, A. Juss.*) in the indigenous systems of medicine, and also of the fact that fairly extensive studies in their chemical constituents carried out by earlier authors [1-6] failed to yield any well-defined products to which these properties could be attributed, it was considered of interest to re-investigate the seeds and the oil for their bitter and sulphur-containing constituents.

The method of isolation evolved after preliminary experiments consisted in the fractional precipitation of the oil with increasing quantities of petroleum ether. Further, the alcoholic extract of the seed-cake was worked up generally following the mild procedure employed by Siddiqui [7,8] in the isolation of Nimbin and other bitter constituents. Thus on removal of the residual oil from the semi-solid alcoholic extractive with petroleum ether, and repeatedly digesting the residue with water, a water-insoluble bitter principle was obtained in the form of a light-brown amorphous powder, which, alongwith the precipitates referred to above, served as starting materials for the present study. Details of the process followed in the isolation of the various constituents are given in the experimental. As a result of this work a series of products, constituents A, B_1, B_2, C and D, have been isolated, which are noted in Table I alongwith data relating to the crystalline products reported by earlier authors which were not, however, obtained in the present procedure of isolation. The table also includes the data for a sulphur containing oily constituent.

Of the two constituents B_1 and B_2, it will be of interest to note that while they have the same melting points and analytical data, one of them is bitter and the other non-bitter. Their optical rotations also vary. In respect of B_1 it has been further noted that it reduces Fehling's solution only after boiling with a few drops of hydrochloric acid, which might indicate a glycosidal structure for it, with $C_{21}H_{26}O_4$ as the aglycone residue after accounting for the sugar moiety. Moreover the presence of a carboxyl group was indicated by the hydroxamic acid test, and the phenol grouping by Millon's reagent and para-nitroaniline. The deep red colour of the solution in 2N sodium hydroxide together with the decolorisation of permanganate solution in the cold indicates a dihydroxy aromatic system. Considering the fact that the glycoside goes into solution in dilute ammonia and sodium carbonate solution indicating the presence of a carboxyl group, it is tentatively, suggested to represent it as follows :—

$$C_{20}H_{23}\begin{cases} OH \\ COOH \\ O\text{-}C_6H_{11}O_5 \end{cases}$$

Further the two crystalline constituents B_1 and B_2 react with 2N sodium hydroxide to give a deep red solution. As all the petroleum ether precipitates and the alcoholic extractive of the cake give the same coloration with 2N sodium hydroxide, it would appear that much larger quantities of the above crystalline constituents occur in the seeds than it has been possible to isolate.

In the course of this work, it was observed that the oil from the seed crop of 1956 yielded a negligible amount of precipitate on treatment with petroleum ether. This recalls the observation of Siddiqui et al. that they were not able to isolate the two crystalline bitters, Nimbin and Nimbinin, from certain fresh samples of solvent-extracted Neem oil and that their yields from various commercial samples varied appreciably.[9] It would thus appear that the nature and yields of the constituents of the Neem seed and oil are liable to variations due to soil and climatic conditions, and other factors connected with the harvesting and storage of the seeds.

Experimental

5.6 kg. of Neem-kernels, pressed out in the oil mill, yielded 1500 cc. of oil and 3.2 kg. of cake.

Treatment of the Oil

The oil (1500 cc.) was successively treated with 375 cc. portions of petroleum ether (b.p. 60-80 °C.). Each time after adding the solvent the oil was warmed to 50 °C. for 15 minutes to flocculate the precipitate formed, and then allowed to stand for 24 hours. The precipitate was then filtered and washed with petroleum ether till free from oil. Any loss of solvent due to evaporation was accounted for in the addition of the next 375 cc. lot of petroleum ether, and the whole process repeated. After 5 such operations no more pre-

cipitate was obtained on further addition of petroleum ether.

However, on standing for a month a substantial quantity of a deposit was obtained. On subsequent addition of petroleum ether and standing no further precipitation was observed. Seven separate fractions were thus obtained, the characterisation of which is given in Table II.

Isolation of constituent A.—Precipitate 2 (1.8 g.) was dissolved in benzene (10 cc.) filtered from a negligible quantity of insoluble material, and the solution concentrated in vacuo to a fluid of thin syrupy consistency. On standing in the cold a crystalline substance was obtained which was sucked off and washed with a few drops of cooled benzene (20 mg.). On recrystallisation from chloroform and alcohol constituent A (8 mg.) was obtained in the form of colourless clusters of tapering rods (Plate I) which melted

Plate I.—Constituent A.

sharply at 95°C. The substance was readily soluble in chloroform, ethyl acetate and benzene, fairly so in alcohol and acetone, almost insoluble in ether and petroleum ether. It was found to contain no sulphur.

Isolation of constituents B_1 and B_2.—Precipitate 6 (23 g.) when treated with alcohol (100 cc.) went into solution. This immediately gave a white precipitate which was filtered, washed with alcohol and dried (2.1 g.). It was dissolved in chloroform (10 cc.) and alcohol (3 cc.) was added to the solution, which was then concentrated in vacuo at 50°C. till faintly turbid. On cooling in ice a crystallisate was obtained. The filtrate and washings from it yielded on concentration in vacuo and cooling, a second crop of crystals. On repeated fractional crystallisation in this manner, B_1 was finally obtained from the tail fractions in the form of wedge-shaped crystals 0.75 g., (Plate II), which darkened at 200°C.

Plate II.—Constituent B_1.

and melted at 307°C. (decomp.). It was readily soluble in chloroform and glacial acetic acid, fairly so in acetone and ethyl acetate, sparingly soluble in alcohol and nearly insoluble in water, benzene, ether and petroleum ether. $[\alpha]^{29}_D = +152°$ in 0.1% solution in alcohol. $[\alpha]^{31.5}_D = +156°$ in 1% chloroform solution.

Calculated for $C_{27}H_{36}O_9$ (505): C, 64.28; H, 7.1; O, 28.57. Found, after drying in vacuo at 50°C.: C, 64.45; H, 7.01; O, 28.33.

B_1 dissolved to a yellow solution on heating in 2N sodium carbonate. In concentrated ammonia it slowly went into solution on standing. With 2N sodium hydroxide, however, it readily yielded a yellow solution, from which the starting material could be got back on immediate acidification. On longer standing the colour deepened to red and B_1 could no more be recovered from the solution. Instead a bright yellow amorphous substance soluble in alcohol was obtained which could not be crystallised. With concentrated hydrochloric acid there was no reaction in the cold. 2N sulphuric acid dissolved the substance slowly on heating, and the concentrated acid gave with it a reddish-coloured solution. Addition of water produced a white flocullent precipitate from the latter. Potassium permanganate solution was decolorised immediately. Fehling's solution was not reduced by an alcoholic solution of B_1. When, however, the latter was boiled with a little hydrochloric acid it readily reduced Fehling's

TABLE I.—CRYSTALLINE OR OTHERWISE CHARACTERISED CONSTITUENTS OF NEEM.

No.	Name	M.P. °C.	B.P. °C.	$[\alpha]_D$	Taste	Molecular formula	Isolated by	Literature
1.	Margosopicrin ..	221-222	—	+163.8°	Bitter	$C_{24}H_{32}O_8$	Watson, Chatterjee and Mukherjee	5
2.	—	Does not melt below 230	—	—325°	Not bitter	$C_{13}H_{24}O_7S_2$		
3.	Neemola (Oil) ..	—	156—158 at 118mm.	—	Not bitter	$C_{15}H_{30}O_3S$	Qudrat-i-Khuda, Ghosh and Mukherjee	6
4.	Nimbin ..	205	—	+170°	Bitter	$C_{28}H_{40}O_8$	Siddiqui	7,8
5.	Nimbinin ..	192	—	—	Bitter	$C_9H_{10}O_3$ (empirical)		
6.	Constituent A ..	95	—	—	Not bitter	—	Present authors	
7.	Constituent B_1 ..	307 (decomp.)	—	+156°	Not bitter	$C_{27}H_{36}O_9$		
8.	Constituent B_2 ..	307 (decomp.)	—	+186.4°	Bitter	$C_9H_{12}O_3$ (empirical)		
9.	Constituent C ..	280 (decomp.)	—	—	Not bitter	—		
10.	Constituent D ..	260 (decomp.)	—	—	Intensely bitter	—		

TABLE II.—FRACTIONAL PRECIPITATION OF NEEM OIL WITH PETROLEUM ETHER.

Amount of Oil=1500 cc.

No. of fractions	Volume of petroleum ether added	Amount of precipitate formed in 24 hrs.	Soluble	Sparingly soluble	Almost insoluble	In dil. alkalies 2N Na_2CO_3, 2N NaOH	In 2N acids, HCl, HNO_3 & H_2SO_4
1	1×375 cc.	2.2 g.	Ethyl Acetate, Chloroform, Acetone, Alcohol	Benzene	Ether, Petroleum ether	Soluble in the cold	Insoluble in the cold
2.	2×375 cc.	1.8 g.	,,	,,	,,	,,	,,
3.	3×375 cc.	2.5 g.	,,	,,	,,	,,	,,
4.	4×375 cc.	2.9 g.	,,	,,	,,	,,	,,
5.	5×375 cc.	9.6 g.	,,	,,	,,	,,	,,
6.	6×375 cc.	23 g. (after standing 1 month)	,,	,,	,,	,,	,,
7.	7×375 cc.	Nil (after standing 6 months)					

solution. Millon's [10] and p-nitroaniline [11] tests for the phenolic group were positive. Test for acid grouping by conversion to hydroxamic acid [12] was also positive.

Plate III.—Constituent B_2.

Isolation of constituent B2.—The first crop of crystals obtained from precipitate 6 was dissolved in chloroform (10 cc.), and alcohol (2 cc.) was added to the solution, which was concentrated in vacuo till a faint turbidity appeared. Repeated recrystallisation of the crystallisate obtained on cooling the concentrate finally yielded B_2 (Plate III) which was bitter to taste, had about the same solubilities as B_1 darkening from 220°C. onwards. It showed $[\alpha]_D^{28} = +186.4°$ in 1 to 2% chloroform solution, as against +156° for B_1, the optical rotation remaining unaffected on further crystallisation.

Calculated for $C_{27}H_{36}O_9$: C, 64.28; H, 7.1; O, 28.57. Found, after drying in vacuo at 50°C. : C, 64.28 ; H, 7.40 ; O, 27.81.

With 2N sodium hydroxide B_2 gave a deep yellow solution from which the original substance could be recovered almost quantitatively on immediate acidification as in the case of B_1.

Precipitate 3 (2.5 g.) yielded through ethyl acetate and ether a crystallisate which appeared under the microscope to have three different types of crystals, one of them corresponding to B_1, and which indefinitely melted from 200 to 300°C. (decomp.).

Precipitate 4 (2.9 g.) and 5 (9.6 g.) when worked up as precipitate 6 yielded 0.05 g. and 0.2 g. respectively of B_1. The mother liquors containing the bulk of the material failed to give any uniform crystalline product.

Treatment of the Cake

Isolation of constituent C.—The cake (3.2 kg.) was percolated 5 times at room temperature with about 5 l. of alcohol for each treatment and the combined percolates were freed of the solvent in vacuo at 50°C. The brown treacly residue was exhaustively extracted with petroleum ether (b.p. 60-80°C.) and then with water. The insoluble product, which formed a yellowish-brown powder failed to yield a crystallisate. The petroleum ether extract gave on standing for about a month at room temperature a cream-coloured precipitate from which a small quantity of B_1 could be crystallised out through alcohol. The ethereal filtrate was freed of the solvent in vacuo at 50°C. and the oily residue allowed to stand for a month, when a precipitate developed, which on crystallisation through alcohol gave white, tasteless, cauliflower-like aggregates of crystals (30 mg.) melting at 280°C. (decomp.) (Plate IV). This substance was sparingly soluble in alcohol and ethyl acetate in the hot, nearly insoluble in other common organic solvents and in water.

Plate IV.—Constituent C.

The substance did not react with 2N sodium carbonate and 2N sodium hydroxide in the cold. With 2N sulphuric, nitric and acetic acids there was no reaction either in the cold or on heating. With concentrated nitric acid it dissolved to a yellowish solution on heating.

Isolation of constituent D.—A sample of freshly expressed oil from the crop of seeds of 1954 (430 cc.) was filtered clear of suspensions and allowed to stand at room temperature (average 20°C.) for a fortnight. The precipitate that developed was filtered off, and the clear oil treated with an equal volume of petroleum ether (b.p.

Plate V.—Constituent D.

60-80°C.) and allowed to stand overnight. The precipitate formed was filtered, washed with petroleum ether till free from oil (2.5 g.). It was then exhaustively extracted at room temperature with water (100 cc.), which left a cream-coloured powder as residue (1.5 g.). The combined filtrates and washings on standing overnight deposited a white crystallisate which was filtered and washed once with cold water. On recrystallisation twice over from alcohol it gave constituent D as feather-shaped crystals (10 mg.), melting at 260°C. (decomp.) (Plate V). It was intensely bitter to taste and was readily soluble in acetone, ethyl and methyl alcohols, fairly so in ethyl acetate and almost insoluble in benzene.

Acknowledgement

The micro-analyses have been carried out by A. Bernhardt, Mikro-analytisches Laboratorium in Max-Planck-Institut fur Kohlenforschung, Mulheim, Ruhr, West Germany.

References

1. Roy and Chatterji, Indian J. Med. Research, **5**, 656 (1917).
2. Dutt and Roy, J. Indian Chem. Soc., **48**, 333T (1929).
3. Sen and Banerjee, J. Indian Chem. Soc., **8**, 773 (1932).
4. Sheshadri et al., Indian J. Pharm., **2**, 201 (1940).
5. Watson, Chatterjee and Mukerjee, J. Soc. Chem. Ind. (London), **42**, 387T (1923).
6. Qudrat-i-Khuda, Ghosh and Mukherjee, J. Indian Chem. Soc., **17**, 189 (1940).
7. Siddiqui, Current Sci. (India), **11**, 278 (1942).
8. Siddiqui, Indian Patent No. 33,649 (1947).
9. Siddiqui, Rao and Mitra, J. Sci. Ind. Research (India), **12B**, 152, (1953).
10. Fiegl, *Spot Tests* (Elsevier Publishing Co., London, 1954) 4th Edition, Vol. II Organic Applications, p. 134.
11. Clarke, *A Handbook of Organic Analysis* (Edwin Arnold and Co., London, 1926), 4th Edition, p. 83.
12. Fiegl, *Spot Tests* (Elsevier Publishing Co., London, 1954), 4th Edition. Vol. II, Organic Applications, p. 140.

DETERMINATION OF OPTIMUM CONDITIONS FOR COOKING AND TREATING INTACT FISH SLICES WITH SPECIAL REFERENCE TO BONE SOFTENING

M. Qudrat-i-Khuda, S. Fazl-i-Rubbi, H. Rahman and N.A. Khan

East Regional Laboratories, Pakistan Council of Scientific and Industrial Research, Tejgaon, Dacca

In continuation of preliminary studies,[1] which showed the feasibility of using chemicals to soften free bones without the fleshy portions of the fish, experiments have been carried out on the effects of different pertinent chemical agents on fish and fish constituents. The flavour, the palatability, the odour and changes in proteins of such flesh are all very significant from the consumer's points of view.[2] These qualities have therefore been determined through the judgements of a panel of tasters and observers, and optimum conditions for cooking fish in the presence of suitable chemicals have been determined on the basis of these tests.

Experimental

Materials.—The chemicals used were mostly guaranteed reagents, while some (citric acid, sodium citrate, borax, etc.) were of B.P. grade. Fresh lemon juice after sterilization was utilized. The fish "foli" (*Notopterus Motopterus*), "ilisha" *Hilsai lisha*, and "putti" (*Barbous puntio*) were purchased from the local markets.

Apparatus and Methods.—Three slices of each fish were cooked in each of several beakers (250 ml.) placed on a sand bath made of a big aluminium pan diameter 14″ and 7.5″ depth with 1″ thick layer of sand. The temperature of the sand bath was maintained at about 110°C. the cooking solutions being at 98° to 100°C. The fish slices were just covered by a chemical solution or by plain water in the control in each beaker for all series of experiments. The fish slices were stirred with glass rods (one in each beaker) at regular intervals during the processes of cooking, which lasted one hour in all cases except those mentioned otherwise. Each series of experiments was conducted on one sand bath under uniform conditions as far as practicable.

From more than forty three chemical agents tried in the preliminary investigation,[1] only sixteen were used in the present experiments. Each chemical treatment was tried in several ways : (a) alone (Table I), (b) together with spices and oil (Table II), (c) the chemical solution being added to fish slices being fried with spices and oil (Table III). Additional experiments were then made following up the above observations : the innocuous borax solution was used in combination with a glycerine solution which further enhances cooking processes[3] (Table IV). In order to confirm some of the previous observations the fish slices immersed in a few chemical solutions were subjected to plain heat treatment for various periods of time (Table V). The main series of experiments were conducted with "foli," an inexpensive fish of complex bony structure, while the costly fish, "illisha," and also a small fish, "putti," were treated only with chemical solutions of favourable concentration (Table VI). In the main series, pieces of "foli" were also treated with extreme concentrations of acetic acid in order to set out the limitations to its use. On the basis of the foregoing experiments, supplemented by further repititions using the optimum concentration of chemicals, the cooking procedures were standardized (Table VII). Attempts were also made to determine the free acidity due to hydrolyzed amino acids in all cooked samples of fish.

The data recorded in Tables I to VII were obtained through an organized panel of tasters and observers. The persons involved in the tasting panel were trained to avoid bias by the decisions of one another. The identity of the sample was not disclosed to any person concerned. The majority opinion was accepted and a specific symbol was used.

Results and Discussion

The results of different experiments are shown in the Tables I—VII, wherein symbolic notations are used, and the chart shows the meaning given to the various symbols in each topic of investigation : turbidity, tolerance, surface effects (visual perception), chemical effects on proteins, degree of bone softening and degree of water retention. A glance through the tabular results reveals several points :

(*a*) Spice and oil eliminate bad odour in certain chemical treatment (cf. items 6 and 7 in Tables I—III).

(*b*) Spice and oil do not interfere with other observations made with chemical substances.

(*c*) Borax provides better cooking media for fish.

(*d*) The feasibility of using chemicals is established and the best conditions (cf. Table VII) are determined.

(*e*) Fish cooked under these optimum

CHART.—EXPLAINING THE SYMBOLS USED IN THE TABLES

Symbols used	Turbidity	Tolerance	Surface effects	Chemical effects on proteins	Degree of bone softening	Degree of water retention	Odour
+	Turbid	Edible	Affected	Mild effects on surface	Mildly malleable	Normal	Normal
++	———	Edible and tasteful	Definite change	Surface mildly hydrolysed	Malleable	Slightly juicy	,,
+++	Very turbid (dark)	Edible and highly tasteful	———	Partially hydrolysed	Fuly malleable	Juicy	,,
++-	———	May be edible	Slight change	Slight hydrolysis	Slightly malleable	Less juicy	,,
+-	Doubtful	May not be edible	———	Doubtful	Doubtful	Doubtful	,,
-	Clear solution (normal)	Hardly edible	No effect (normal)	No change (normal)	No effect	Slightly dehydrated	Not good
--	———	Not edible	———	———	———	Dehydrated	,,
---	———	Obnoxious	———	———	———	Very much dehydrated and granular	,,

TABLE I.—HEAT TREATMENT OF INTACT FISH AT 98°—100°C.

No. of test	Solutions used	Observations (after) one hour					
		Turbidity	Tolerance	Chemical effects on proteins	Degree of bone softening	Degree of water retention	Odour
1	Citric acid 1.25%	+	+	+(-)	+	+	+
2	,, ,, 2.5%	+-	-	++	+	+	+
3	Acetic acid 1%	++	+	+	+	+	+
4	,, ,, 1.5%	+	-	+	+	+	+
5	,, ,, 2%	+	--	+	+	-	+
6	,, ,, 2.5%	-	---	++	+	-	-
7	,, ,, 3%	-	---	+++	++	-	--
8	Acetic acid 2% Sodium citrate 1% ,, acetate 1%	++	---	-	+	-	+
9	Citric acid 2.5% Borax ,, 0.2%	-	---	+	+	+	+
10	Citric acid 2.5% Borax ,, 0.2%	-	+	+	+	+	+
11	Sodium phosphate 1% o-Phosphoric acid (2 drops in 100 ml.)	+++	++-	+	-	-	+
12	Borax 0.5%	-	+++	+	+-	+	+
13	,, 1%	-	+++	++	+-	+	+
14	,, 3%	-	+++	+++	+-	++	+
15	,, 5%	-	++	+++	++	++	+
16	Lemon juice (fresh)	-	-	+	-	+	+
17	Water as control	-	+	-	-	+	+

TABLE II.—COOKING OF INTACT FISH WITH SPICES AND OIL.

No. of tests	Observations (spices and oil) one hour				
	Tolerance	Chemical effects on proteins	Degree of bone softening	Degree of water retention	Odour
1	+ + −	−	−	+	+
2	+	+	+	+	+
3	+	+	−	+	+
4	+	+	+	+	+
5	+	+	+	+ −	+
6	+	+	+	+	+
7	+	+ +	+	+	+ −
8	+	+ −	+ +	+ −	+
9	+	+	+ −	−	+
10	+	+ +	+	−	+
11	+ −	+ −	+ −	−	+
12	+ +	−	+	+ + +	+
13	+ +	−	+	+ +	+
14	+ +	−	+ +	+ + −	+
15	+ +	−	+ +	+	+
16	+	+ +	+ −	−	+
17	+	−	−	+	+

TABLE III.—COOKING OF INTACT FISH WITH SPICES FRIED IN OIL.

No. of tests	Observations (spices and oil) one hour				
	Tolerance	Chemical effects on proteins	Degree of bone softening	Degree of water retention	Odour
1	+	+	−	+ −	+
2	+	+	+	+ −	+
3	+	+ + −	−	+ −	+
4	+	+ + −	+	+ −	+
5	+	+ + −	+	+ −	+
6	+	+ + −	+	−	+
7	+ −	+ +	+	−	+
8	+	+	+	+	+
9	+	+	+ + −	+	+
10	+	+ −	+ + −	+	+
11	+	−	+ + −	+ + −	+
12	+ + +	+ −	+	+	+
13	+ +	+ −	+	+	+
14	+ +	+	+	+	+
15	+ +	+	+ + −	+ +	+
15a (Borax 10%)	+ +	+ +	+ + −	+ +	+
16	+	+ +	+	+ −	+
17	+	−	−	+	+

TABLE IV.—SPECIAL HEAT TREATMENT OF INTACT FISH AT 98°—100°C.

No. of test	Solutions used	Observation after one hour				
		Tolerance	Chemical effects on proteins	Degree of bone softening	Degree of water retention	Odour
1	Borax 3% + Glycerine	+ +	+	+	+ +	+
2	Borax 5% + Glycerine	+ +	+ +	+	+ +	+
3	Borax 10% + Glycerine	+ +	+ +	+	+ +	+
4	Water 200 ml. Glycerine 0.5 ml.	+	−	−	+	+
5	Water 100 ml.	+	−	−	+	+

TABLE V.—HEAT TREATMENT OF INTACT FISH UNDER DIFFERENT CONDITIONS. (TEMPERATURE OF SOLUTIONS : 98°—100°C.)

No. of test	Solutions used	Observations					
		Tolerance	Chemical effects on proteins	Surface effect	Degree of bone softening	Degree of water retention	Duration
1	Citric acid 2.5%	+ +	+	+	+	+	1 hr.
2	Citric acid 2.5% Borax 0.2%	+	+ +	+ +	+ +	+	,,
3	Acetic acid 4%	+ −	+ +	+ +	+ +	−	,,
4	Borax 2%	+	−	−	−	+	,,
5	,, 4%	+	+	+	+	+ +	,,
6	,, 5%	+	+	+	+	+ +	,,
7	,, 10%	+	+	+	+	+ + +	,,
8	Water	+ −	−	−	−	Normal	,,
1	Citric acid 2.5%	+	+ +	+	+	−	2 hrs.
2	Citric acid 2.5% Borax 0.2%	−	+ +	+ +	+	−	,,
3	Acetic acid 4%	+	+ +	+ +	+ +	−	,,
4	Borax 2%	+	+	+	+	+	,,
5	,, 4%	+	+	+	+	+	,,
6	,, 5%	+	+	+	+	+	,,
7	,, 10%	+	+	+	+	+	,,
8	Water	+ −	−	−	−	Normal	,,
1	Citric acid 2.5%	+	+ +	+ +	+ +	−	3 hrs.
2	Citric acid 2.5% Borax 0.2%	+	+	+	+	−	,,
3	Acetic acid 4%	+	+ +	+ +	+ +	−	,,
4	Borax 2%	+	+	+	+ −	+ +	,,
5	,, 4%	+	+	+	+ −	+ +	,,
6	,, 5%	+	+	+	+ −	+ +	,,
7	,, 10%	+	+	+	+ −	+ +	,,
8	Water	+	−	−	−	Normal	,,

Table VI.—Treatment of Intact Fish Slices under Optimum Conditions.

Name of fish in pieces	Solutions used	Tolerance	Chemical effects on proteins	Degree of bone softening	Degree of water retention	Odour
Illisha	Acetic acid 1.0% (a)	+	–	+	–	+
	Citric 1.5% (b)	+	–	+ +	+	+
	Borax 5.0% (c)	+	+ +	+ +	+ +	+
Putti	a	+	–	+	–	+
	b	+	–	+ +	+	+
	c	+	+	+ +	+ +	+
Foli	Acetic acid 10.0%	–	+ + +	+ + +	– – –	–

Table VII.—Optimum Conditions for Cooking Intact Fish Slices with Chemicals in Presence of Spices.

Chemicals	Range of concentrations	Duration hrs.	Temperature	Heating arrangements
Acetic acid	0.5 — 2.0%	½ — 1½	95 — 98°C.	Water or steam bath
Citric	0.5 — 3.0%	1 — 2	110 — 120°C.	Sand bath
Borax	3.0 — 7.5%	1 — 2	110 — 120°C.	,, ,,
Lemon juice	Same as in fruits	1	105 — 110°C.	,, ,,
Water (control)	110%	1 — 1½	95 — 98°C.	Open burner or electric heater (controlled)

conditions show several desirable features. (cf. Table VI).

These observations point the way to future progress in work on fish bones softening in the intact fish through suitably adapted chemical treatment. The hydrolysis of proteins to amino acids during such cooking process was not significant enough to be detected by the ordinary titration methods. It therefore appears that the fundamental aspects of protein loss, nutritive degradation, toxicity and others that might be caused by chemical treatments, have been essentially controlled. Further work is in progress and the results will be published later.

Acknowledgement

The authors wish to thank Dr. Salimuzzaman Siddiqui, Director, Pakistan Council of Scientific and Industrial Research, Karachi, for his helpful suggestions during the progress of the work.

References

1. M.Q. Khuda, S. Fazl-i-Rubbi, and N.A. Khan, paper read at the tenth Pakistan Science Conference, March, 1958.
2. Md. Sweetman and I. Mackeller, *Food Selection and Preparation*, (John Wiley and Sons, Inc., New York, 1954), 4th edition.

STUDIES ON SHARK LIVER OIL

Part II.—Recovery of Vitamin 'A' by Different Digestion Methods

S. Maqsood Ali, S. Abdul Haq and S. Mahdihassan

Central Laboratories, Pakistan Council of Scientific and Industrial Research, Karachi

Introduction

Fish livers have been classified[1] according to their oil and vitamin 'A' contents, as (i) low potency, high oil-content livers, (ii) high potency, low oil-content livers and (iii) high potency, high oil-content livers. Class (i) livers are generally digested with steam or simply hot water, while for those belonging to class (ii) and (iii) the digestion process is modified, the purpose in each case being the optimum extraction of both oil and vitamin A. Simple digestion of the livers of type (ii) and (iii) with hot water or steam always leaves a portion of the oil and the vitamin firmly bound in the tissue, and complete digestion to a liquid or semi-liquid state becomes imperative for total recovery of oil and vitamin A.

Steam, alkali, and enzyme digestion, and also solvent extraction have been employed by various workers [2,3,4], on different fishes, and most of these processes are covered by patents. Keeping in view the efficient exploitation of the vitamin A resources of Pakistan, five possible methods have been studied in the present investigation in order to evolve a process which could be commerically feasible for the maximum extraction of oil and vitamin A from indigenous sharks.

Experimental

Liver of two species of sharks, *P. cuspidatus* and *S. blochii*, were procured from Khadda Market, Karachi. These livers were washed, passed through a meat chopper and digested in the following manner:—

(*i*) *Water treatment*: To 500 g. of liver 500 ml. of tap water was added, followed by heating at 80-85 °C. for 45 minutes with occasional stirring. The oil was separated by centrifugation and filtered hot through anhydrous sodium sulphate into a tared flask.

(*ii*) *Alkali digestion*: (2% *alkali*): To 500 g. of liver in 500 ml. of water, 15 ml. of saturated aqueous sodium hydroxide (equivalent to 10 g. of NaOH) was added with constant stirring. The mass was digested at 75-80 °C. for about 20 minutes, whereupon all the tissue had been dissolved and an emulsion obtained. The excess aqueous phase was removed in a separating funnel and the emulsion was broken by the addition of an equal volume of 5% hot sodium chloride solution. The oil thus liberated wa centrifuged, dried and filtered into a tared flask.

(*iii*) *Papain digestion* (1% *Papain*): 5g. of crude papain was suspended in 100 g. of warm water at 50 °C. This suspension was added to 500 g. of liver, the whole was diluted with 400 ml. of water, and was then digested at 55 °C. for 3 hours, with occasional stirring. When the digestion was complete, the oil was recovered in the usual manner.

(*iv*) *Steam digestion*: Steam was injected for about 30 minutes into a two litre flask containing 500 g. of liver and 500 ml. of water. The liberated oil was collected as usual.

(*v*) *Ether extraction*: 500 g. of liver were ground with about 100 g. of anhydrous sodium sulphate and repeatedly extracted with ether, until a portion of the extract left no residue on evaporation. The oil was recovered from the extract by distilling off the ether.

The acidity of the oil was determined in terms of oleic acid, and its vitamin A content was estimated spectrophotometrically.[5] The results are tabulated in Tables 1 and 2.

Discussion

The tables show that the maximum yield of oil is obtained with ether extraction, while alkali and papain digestion come fairly close to this. Water and steam both give low yields, amounting to about two-thirds of the maximum. On the basis of recovery of vitamin A, ether digestion again yields the maximum quantity, but alkali digestion is now equally efficient and is closely followed by papain. Water extraction again gives the lowest yield and steaming is slightly better, the difference being more conspicuous with medium oil content liver. Another noteworthy feature is that alkali and papain digestion yield a more potent oil than that obtained by water, steam, or ether extractions.

Alkali, as expected, yields oil with the lowest

TABLE 1.—ANALYSIS OF OIL EXTRACTED FROM HIGH OIL CONTENT LIVER (*P. Cuspidatus*).

Extraction method	Oil from ½ kg. liver in grams	Yeild of oil % age	Acidity (as oleic acid) % age	Vit. A (I. U. per g. of oil)	Units of Vit. A per kg. liver in million units
Water ..	190	38	0.56	12,300	4.7
2 % Alkali ..	250	50	0.08	15,700	7.8
1 % Crude papain	240	48	1.80	14,900	7.2
Steam ..	190	38	0.73	14,300	5.4
Ether ..	280	56	0.76	13,800	7.7

TABLE 2.—ANALYSIS OF OIL EXTRACTED FROM MEDIUM OIL CONTENT LIVER (*S. blochii*).

Extraction method	Oil from ½ kg. liver in grams	Yield of oil % age	Acidity (as oleic acid) % age	Vit. A (I.U. per g. of oil)	Units of Vit. A per kg. liver in million units
Water ..	101	20	1.47	44,300	8.9
2 % Alkali ..	150	30	0.28	62,600	18.8
1 % Crude papain	120	24	1.81	62,700	15.0
Steam ..	110	22	1.46	51,902	11.4
Ether ..	170	34	1.50	55,300	18.8

acidity, while maximum acidity is obtained with papain digestion, due to the action of fat-splitting enzymes. It is to be noted that the oil obtained by the four processes other than alkali digestion needs neutralisation before being used.

From the above results, it follows that the best extraction method for indigenous shark livers is alkali digestion. This conclusion is in agreement with the experiments of Hartman[6] and others.[3,4] However, when the residual liquid is to be further processed for obtaining the anti-anaemic factor and other water soluble vitamins, papain digestion will be the most suitable.

References

1. U.S. Dept. of Interior, Fishery leaflet No. 233.
2. S. Tachino and H. Tankana, Bull. Japan Soc. Sci. Fisheries, **19**, 533-6 (1953).
3. P.N. Sarangdhar, J. Sci. Ind. Research (India), **6B**, 180-5 (1947).
4. E.J. Ferguson Wood, J. Council Sci. Ind. Research (Australia) **14**, 311-14 (1942).
5. Vitamin Assays (Interscience Publishers, Inc., New York, (1951) Second Edition pp. 36-39.
6. L. Hartman, J. Am. Oil Chemists' Soc. **27**, 409, (1950).

COBALT CONTENT OF EAST PAKISTAN FISHES*

MOHAMMAD ABUL HUSSAIN SHARIF † AND KAMALUDDIN AHMED

Biochemistry and Nutrition Department, University of Dacca, Dacca, East Pakistan

Introduction

The occurrence and distribution of cobalt in biological materials has attracted the attention of several investigators during the last 30 years. Bertrand and Mokragnatz[1] as early as 1922 found cobalt and nickel to occur in plants and later Bertrand and Macheboeuf[2] detected these elements in most organs of man and other mamals, birds, fishes and marine animals which they examined. The first evidence that cobalt is an essential nutrient for mammals came from the comprehensive study of Filmer and Underwood[3] who traced the prophylactic value of an iron-free extract of limonite (Fe_2O_3, H_2O) (known to be therapeutically active), against what was known as "Denmark disease" or "Enzootic marasmus" in the farm animals of Western Australia. The similarity of this disease to "Bush sickness" in New Zealand, "salt sick" in Florida, "pine" in Scotland and "Nakuritis" in Kenya was pointed out by Filmer. Askew et al.[4] later confirmed that the deficiency of cobalt in soils that served as the pasture land for these animals was responsible for the disease.

With the discovery of vitamin B_{12}, the anti-pernicious anaemia factor, which contains one atom of cobalt per molecule,[5] cobalt has assumed tremendous importance in human hematopoeisis. Since fishes are one of the sources of vitamin B_{12}, and since they constitute the main source of animal proteins for the bulk of the population of East Pakistan, considerable importance attaches to a survey of the cobalt content of these fishes. This is particularly so because there are no available data for the cobalt or other trace element contents of East Pakistan fishes. The present paper describes such a survey, on the basis of which an attempt has been made to find out the regions where cobalt rich fishes are available in plenty.

Materials and Methods

For the colorimetric estimation of cobalt in fishes, the method used by Kidson and Askew[6] was followed, the procedure adopted being briefly described as follows :—

TABLE 1.—THE COBALT CONTENT OF THE IMPORTANT EAST PAKISTAN FISHES.

Zoological name	Local name	Sources	Cobalt content p.p.m.
Labeo rohita	Rohit	Ponds & lakes	0.42
Catla catla	Katla	,,	0.46
Labeo calbasu	Kalibahus	,,	0.21
Clarius batrachus	Magur	,,	0.32
Heteropneustes fossiles	Singil	,,	0.38
Anabas testudineus	Kaii	,,	0.42
Wallagoni attu	Boal	Rivers & canals	0.35
Mystus cavasius	Pungas	,,	0.21
Mystus vittatus	Tangra	,,	0.80
Hilsa ilisha	Hilsha	Rivers & bays	0.38
Ompok bimaculatus	Shilan	Rivers	0.86
Notopterus notopterus	Foli	Ponds & lakes	0.29
Barbus sarana	Puti	Canals	0.27
Palaemon carcinus	Chingri (big)	Rivers & canals	0.50
Palaemon carcinus	Chingri (small)	,,	0.51
Mastacembelus armatus (*Lacep*)	Baim	Canals	0.49
Glossogobinus giuris	Bele	,,	0.53
Ophicephalus marulius	Qajal	Ponds	0.21
Ophicephalus sonnerati	Shouli	,,	0.25
Serranus sonnerati	Bhetki	Ponds, rivers & canals	0.29

Note.—The values of cobalt content represent the average of two determinations and are on the basis of dry weight of whole fish.

* This paper is based on M. Sc. dissertation presented at the University of Dacca by M.A.H. Sharif in 1951.

† Now at Central Laboratories, Pakistan Council of Scientific and Industrial Research, Karachi.

The particular fish under investigations were dried at a constant temperature of 110°C. for 48 hours and from the dried material 10 g. of sample was taken and mixed in a silica basin with 30 ml. of 7.5N nitric acid. After ignition, the basin was put into a muffle furnace, where it was heated to a dull red heat for 12 hours. The ash was moistened with 5-10 ml. of the same acid, dried and ignited again. The process was repeated a third time, after which the ash was taken up with 30 ml. of 4N hydrochloric acid and heated gently for 20 minutes. The solution was filtered into a Pyrex dish and the residue washed three times with hot water. The filtered solution was evaporated nearly to dryness and then taken up in 10 ml. of water.

10 ml. of a standard comparison aqueous solution of cobalt chloride and a "blank" consisting of 10 ml. of distilled water were also prepared at this stage, and from here onwards the standard and blank solutions were treated in the same manner as the unknown. After the addition of 1 ml. of 4N hydrochloric acid and eight drops of 7.5N nitric acid, the solution was transferred to a 100 ml. Erlenmeyer flask and boiled for a few minutes to oxidise any reducing substance and then cooled. Exactly 2 ml. of 0.1% nitroso R.-salt in water and 2 g. of sodium acetate were then added and the solution warmed to 70°C. to develop the characteristic colour fully. In order to stabilise this colour, the following procedure was adopted : 10% potassium hydroxide was gradually added with constant shaking until the solution was just alkaline to phenolphthalein (0.2% in alcohol), after which sufficient 0.5N hydrochloric acid was added to make the solution just acid. 5 ml. of 7.5N nitric acid were then added (to decompose the complexes of most of the other heavy metals that may be present), the solution boiled for 2 minutes and then cooled under a tap. The volume of the solution was made upto 25 ml. and filtered in a dark room.

The characteristic red colour developed due to the cobaltic ions was matched against the cobalt standards in a colorimeter with a green filter, the "blank" water sample being used to set the zero of the instrument. Table 1 shows the results obtained for twenty different varieties of fish. The variation between successive determination was always less than 0.1 p.p.m.

Discussion

It appears from the table that the cobalt content of these fishes varies from 0.21 to 0.86 p.p.m., the lowest quantities being found in fishes of the types *Labeo calbasu*, *Ophicephalus marulius*, *Sarranus sonnerati*, while the highest occur in fishes of the varieties *Ompok bimaculatus*, *Mystus vittatus*. Moreover, it is observed that the fishes from ponds, lakes etc., generally have a low cobalt content while those from rivers and canals contain relatively large quantities of this element. Table 1 lists ten varieties of fish from each of the two types of sources, and we find that the mean cobalt content of the fish from lakes and ponds is 0.33 p.p.m., while that of river and canal fish is 0.49 p.p.m., *i.e.*, 50% higher. Our results are the only ones available for East Pakistan.

References

1. G. Bertrand, and M. Mokragnatz, C.R. Acad. Sci., **175**, 458 (1922).
2. G. Bertrand and M. Macheboef, Ibid., **180**, 1380 (1925).
3. Underwood and Filmer. Austral. Vet. J., **11**, 89 (1935).
4. Askew and Dixon. New Zealand. J. Sci. Technol., **19**, 317 (1937).
5. Hodgkin et al., Nature, **176**, 325 (1955).
6. E.B. Kidson and H.O. Askew. New Zealand Sci. Technol., **21B**, 178-189, (1940).

ESTIMATION OF VITAMIN B12 BY COLORIMETRIC DETERMINATION OF COBALT

MOHAMMAD ABUL HUSSAIN SHARIF

Central Laboratories, Pakistan Council of Scientific and Industrial Research, Karachi

Introduction

Of the various methods that have been suggested for assaying vitamin B_{12}, the microbiological method is the one most frequently used. Another biological method using laboratory animals is also employed, but this is laborious and time consuming. In fact the microbiological method suffers from several drawbacks, among which may be mentioned the variable activities displayed by different naturally occurring forms of vitamin B_{12}. The fact that desoxyribosides and some reducing agents also permit growth of the microbes under the same condition as vitamin B_{12}, introduces another complication on the biological methods of estimation. Several specific chemical and physico-chemical methods for the estimation of the vitamin have therefore been developed. Because of the characteristic ultra-violet absorption spectrum of vitamin B_{12}, the spectrophotometric method was developed early on, and the ratio of the absorbency at wave lengths 360 m μ to that at 548 m μ (theoretical value 3.24) is used in the absence of interfering substances as a measure of the vitamin B_{12} present. The colorimetric methods include (i) the cyanide assay,[2] in which the cyanide from cyano-cobalamin is liberated by aeration of an aqueous solution at pH 5, (ii) the cyanide complex method[3] based on the difference between the visible spectrum of cyano-cobalamin and its di-cyanide complex, this difference being maximal at 582 m μ with E_{1cm} 1%=54 and (iv) the measurement of 5,6-dimethyl-benzimidazole, an hydrolysis product of vitamin B_{12},[4] the quantitative liberation of 5,6-dimethylbenzimidazole being attained by the action of 0.1N hydrochloric acid at 120°C. for 4 to 16 hours. Other methods are (a) the countercurrent distribution assay, in which use is made of the distribution of vitamin B_{12} in the water-benzyl alcohol system,[5] subject to the absence of other coloured substances, and (b) the radio-active tracer method depending upon the radioactive tracer dilution principle and using cyanocobalamin with labelled cobalt,[6] which is especially applicable to mixtures containing very small amounts of vitamin B_{12}, where quantitative isolation of the vitamin is not possible.

From the foregoing, it is seen that the chemical methods thus far suggested are somewhat lengthy and are not always applicable to the raw materials that form sources of vitamin B_{12}. There is therefore a need for a chemical method that is short and simple, and is applicable equally well to the different forms of vitamin B_{12} and to the food stuffs and biological materials that contain the vitamin. A method fulfilling these requirements is described below and experimental data on its application are given. An accurate determination of the cobalt content of a sample enables the quantity of the vitamin to be deduced from its formula, $C_{63}H_{88}N_{14}O_{14}CoP$,[7,8] because, apart from the vitamin B12 group, no cobalt-containing organic compounds have yet been found in nature. One gram of vitamin B_{12} contains 0.0435 gm. of cobalt, and therefore, the cobalt content of a sample divided by 0.0435 (*i.e.*, multiplied by 23.0) can be taken as a measure of the vitamin B_{12}.

Experimental

The method was tested with "Cytacon" vitamin B_{12} syrup (Glaxo), and "Campolon" liquid extract of liver (Bayer), and a further test was also made on the quantitative character of the recovery of cobalt from the syrup and the extract.

In each case the cobalt content of the vitamin was determined by the nitroso R-salt (1-nitroso, 2-napthol, 3,6-disodium sulphonate) reagent according to the procedure described by Kidson and Askew[9] and previously used by the author for a survey of the cobalt content of East Pakistan fish.[10].

1. "*Cytacon*" *vitamin B_{12} made by Glaxo Laboratories*. This liquid syrup of vitamin B_{12}, meant for oral administration, is stated by the manufacturers to contain 7 micrograms of crystalline vitamin B_{12} per ml. 10 ml. portions of this syrup were taken in a platinum dish and processed as described in references (9) and (10), so as to obtain a cobalt complex of the nitroso R-salt which has a characteristic colour. The solutions were matched with freshly prepared cobalt standard in a Hilger colorimeter, using the complimentary colour at wave length 550 m μ.

This procedure was repeated with 20 ml. and 30 ml. portions of the syrup and the results are shown in Table I.

(2) "*Campolon*" *liver extract manufactured by Bayer*. This injectable extract of liver contains 30

TABLE 1.—TEST OF THE METHOD WITH "CYTACON VITAMIN B_{12} SYRUP.

No. of sample	Volume of syrup taken ml.	Nominal values (micrograms)		Values found by the suggested method (micrograms)		Error %	Mean error %
		Cobalt content	Vit. B_{12} content	Cobalt	Vit. B_{12}		
1	10	3.044	70	3.25	74.7	6.7	3.2
2	,,	,,	,,	3.0	69.0	1.4	
3	,,	,,	,,	3.0	69.0	1.4	
				Mean=	70.9		
1	20	6.088	140	6.09	140.1	0.0	1.4
2	,,	,,	,,	6.0	138.0	—1.4	
3	,,	,,	,,	6.25	143.0	2.6	
4	,,	,,	,,	6.0	138.0	—1.4	
5	,,	,,	,,	,,	,,	,,	
6	,,	,,	,,	,,	,,	,,	
				Mean=	139.3		
1	30	9.132	210	9.0	207.0	—1.4	1.3
2	,,	,,	,,	9.2	211.6	0.7	
3	,,	,,	,,	9.0	207.0	—1.4	
4	,,	,,	,,	,,	,,	,,	
5	,,	,,	,,	,,	,,	,,	
6	,,	,,	,,	,,	,,	,,	
				Mean=	207.8		

micrograms of vitamin B_{12} per ml. The determinations of cobalt in this sample were carried out by two different processes. First it was determined as in the case of Cytacon syrup, and it was found that the values obtained were consistently higher than the nominal contents. So it was conjectured that iron, which is abundant in liver tissues, was interfering with the cobalt determination. Therefore, the iron was removed by coverting it into ferric chloride and extracting with ether.[11] The amount of cobalt determined from the aqueous extract got after removal of this iron was found to agree to within 3% with the theoretical values, as shown in Table 2.

TABLE 2.—TESTING OF THE METHOD WITH BAYER'S LIVER EXTRACT.

	No. of Sample	Volume of liver extract (ml.)	Nominal values (microgram)		Values found by the suggested method (microgram)		Error %	Mean error %
			Cobalt present	B_{12} present	Cobalt	B_{12}		
Before removal of iron	1	3	3·903	90.0	4.6	105.8	17·5	
	2	,,	,,	,,	4·7	108.1	20.1	19
	3	,,	,,	,,	,,	,,	,,	
	4	,,	,,	,,	,,	,,	,,	
						Mean=107·5		
After removal of iron	1	3	3·903	90.0	4.0	92.0	2.2	
	2	,,	,,	,,	,,	,,	,,	3·3
	3	,,	,,	,,	,,	,,	,,	
	4	,,	,,	,,	,,	,,	,,	
	5	,,	,,	,,	4·25	97·7	8.6	
	6	,,	,,	,,	4.0	92.2	2.2	
						Mean=93.0		

(3) *Cobalt recovery from the syrup and the liver extract.* To study the recovery of cobalt from the syrup and also from the liver extract, some experiments were carried out by adding calculated amounts of cobalt in the form of cobalt chloride solution to both the Cytacon vitamin B_{12} syrup and the injectable liver extract. The samples were digested and ashed as before, and analysed for cobalt contents. The results are presented in Table 3 and indicate nearly 100% recovery.

Discussion

From Table 1, it is observed that the method gives quite satisfactory results with 20 ml. and 30 ml. of vitamin B_{12}, the error being less than 1.5%. With 10 ml. samples, however, the error rises to 3%, which is understandable because we now have a total of only 3.044 microgram of cobalt. For a 2% accuracy of determination, 4 micrograms of cobalt *i.e.*, about 100 micrograms of vitamin B_{12}, are required, which fact is further substantiated by our results in Table 2, in which are presented the analyses of samples of injectable liver extracts prepared by Bayer under the trade name of "*Campolon.*" In this case, it was necessary to remove the iron before determining the cobalt content as otherwise the iron interferes seriously and raises the estimated cobalt content by nearly 20%. After the removal of iron, however, the method again gives quite satisfactory results as shown by Table 2. The experimental values, however, still tend to be high by 2-3%, which may be due to incomplete removal of the iron or the presence of traces of copper.

Table 3 shows the recovery of cobalt after addition to the Cytacon B_{12} syrup and to the liver extract. Calculated amounts of cobalt in the form of cobalt chloride solution were added to the samples before analysis and the total cobalt contents were estimated as before. From the table it is evident that, except for two or three cases, the error is always less than 2%. Here again the experimental estimates for the liver extract are seen to be higher by about 4%.

TABLE 3.—RECOVERY OF COBALT FROM THE SAMPLES.

	No. of sample	Volume of samples (ml.)	Cobalt content of the sample (microgram)	Amount of cobalt added (microgram)	Total amount of cobalt present (microgram)	Total amount of cobalt obtained by the method (microgram)	Recovery
Cytocon syrup	1	10	3·044	3·0	6·044	6·0	98·5
	2	,,	,,	,,	,,	,,	,,
	3	,,	,,	,,	,,	,,	,,
	4	,,	,,	,,	,,	,,	,,
	5	,,	,,	,,	,,	,,	,,
	6	,,	,,	,,	,,	5·75	90·2
	7	,,	,,	,,	,,	6·0	98·3
	8	,,	,,	,,	,,	5·75	90·2
							Mean=96·4
Liver extract	1	3	3·913	5·0	8·913	9·25	106·7
	2	,,	,,	,,	,,	9·0	101·7
	3	,,	,,	,,	,,	9·0	101·7
	4	,,	,,	,,	,,	8·75	96·7
	5	,,	,,	,,	,,	8·75	96·7
	6	,,	,,	,,	,,	9·0	101·7
							Mean=100·9

The above experiments show that the method suggested can be used for the estimation of vitamin B_{12} to an accuracy of 2%, provided adequate care is exercised against interfering elements. The method has further been tested with two natural sources of vitamin B_{12}, namely beef liver and shark liver. The results obtained by this chemical method, 253 microgram and 191 microgram, respectively, per 100 g. of fresh tissue, are in satisfactory agreement with the recorded values[12,13] obtained by microbiological methods.

A possible source of error in the method when applied to raw biological materials is that there may occur some cobalt which is not a part of vitamin B_{12}. This extraneous cobalt will also be included in the estimate made by this method, and will thereby produce higher estimates for vitamin B_{12}. However, for such materials we can work out an emperical correction factor by comparison with the value obtained from microbiological assay. This is being investigated further, but inspite of this drawback, the method will, in any case, give us a good approximation to the vitamin B_{12} content of biological materials and should be valuable for laboratories that are not equipped with microbiological techniques. For pure vitamin B_{12} and its preparations the method is entirely satisfactory, being accurate to 2%.

Acknowledgement

I am thankful to Dr. S. Mahdihassan, Head of Biochemical Research Division, for his valuable suggestions and encouragement during the work.

References

1. Fantes et al., Proc. Roy. Soc. (London), **B136**, 592 (1949).
2. G.E. Boxer and J.C. Richards, Arch. Biochem. **30**, 372, 382, 392 (1951).
3. G.O. Rudkin Jr. and R.J. Taylor, Anal. Chem. **24**, 1155 (1952).
4. G.E. Boxer and J.C. Richards, Arch. Biochem., **29**, 75 (1950).
5. J.G. Heathcote, J. Pharm. and Pharmacol. **4**, 641 (1952); Chaiet et al., Science, **III**, 601, (1950).
6. Bacher et al., Anal. Chem., **26**, 1146 (1954).
7. Hodgkin et al., Nature **178**, 64 (1946).
8. A.W. Johnson and A. Todd, Endeavour, **15**, 29 (1956).
9. E.B. Kidson and H.O. Askew, New Zealand Sci. Technol. **21B**, 178, (1940).
10. M.A.H. Sharif and K. Ahmad, Pakistan J. Sci. Ind. Research, this issue, p. 158.
11. K. Ahmad and M.A.H. Sharif, Pakistan J. Sci. Research **5**, 119, (1953).
12. T.F. Zucker and L.M. Zucker, *Vitamins and Hormones* (Academic Press, New York, 1950) Vol. VIII, p. 24.
13. B. Truscott and P.L. Hoagland, Can. J. Biochem. and Physiol, **34**, 191 (1956).

THE PUPA AND LARVA OF EUBLEMMA SCITULA

S. MAHDIHASSAN

Central Laboratories, Pakistan Council of Scientific and Industrial Research, Karachi

The lac insects of the continent of India and Pakistan are subject to attack by the caterpillars of *Eublemma amabilis*. There are two other species related to this moth, *viz*. *E. coccidiphaga* and *E. scitula* which are also important. On *E. coccidiphaga* an article has already been published.[1] *E. scitula*, is quite a common parasite of most scale insects but usually not of lac. However, with Sind lac it appears to be the most common of the three species. It has emerged from *Lakshadia sindica* found naturally on *Zizyphus jujuba* in the city of Karachi.

It might be imagined that *E. scitula*, already found on other scale insects, could conveniently infect lac on trees growing nearby. However, during the author's considerable experience of similar lac on trees within the municipal limits of cities like Bangalore, Hyderabad Deccan, Bombay, Madras, and New Delhi, no specimen of *E. scitula* was ever found. Karachi, with its species, *Lakshadia sindica*, stands as an exception to this rule. Near Hyderabad Sind, lac is cultivated on *Acacia arabica*, and in such material again, *E. scitula* was found in greater numbers than *E. amabilis*.

The egg of *E. scitula* has been previously illustrated.[1] The larva builds over itself a case with which it moves about well protected. This covering or shield is held by the serrated rows on the dorsal surface of the hind segment seen at the end of the body in Fig. 1. It shows the dorsal view with the posterior end towards the reader. The scale indicates the actual size of the adult caterpillar freed from its covering. Fig. 2 shows the same caterpillar in profile with the posterior end to the left of the reader and the last segment with the serrated portion in profile. Fig. 3 gives the ventral aspect of the same caterpillar; two pairs of serrated rows enable the caterpillar to clamp fast to the twig or to the encrustation of lac, so much so that it is not easy even for a man to dislodge the caterpillar easily from its place. It is thus impossible for the severest winds to disturb it. In all, there are three pairs of serrated rows serving like pseudo-legs, two pairs on the ventral surface, seen in Fig. 3, and one on the last segment as in Figs. 1 and 2, serving to hold the shield or cover under which it moves like a tortoise. This dorsal pair of serrated rows is shown further enlarged in Fig. 4.

The larva before it pupates builds a tough parchment-like cocoon which is by no means easy to tear. From such an envelope, a pupa was removed which gave Fig. 5 as its dorsal view, Fig. 6 as its ventral, and Fig. 7 as its side-view.

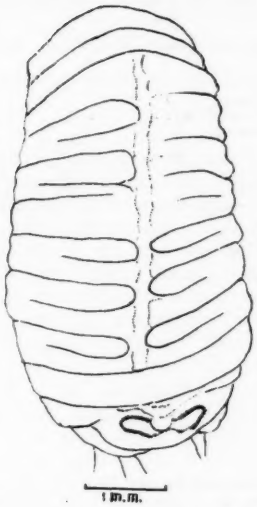


Fig. 1.—*Eublemma scitula*, full grown larva, dorsal view, with the posterior end towards the reader. Note the serrated row on either side of the last segment which holds a shield-like cover to protect itself.

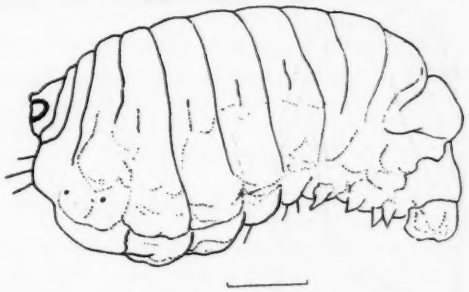


Fig. 2.—*E. scitula*, larva, as in Fig. 1, side-view.

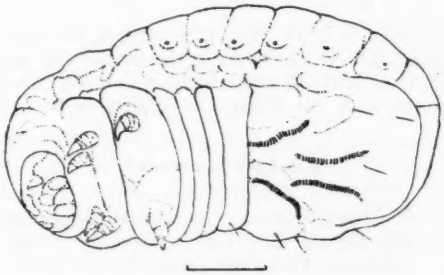


Fig. 3.—*E. scitula*, larva, ventral view. There are two serrated rows used as pseudo-legs.

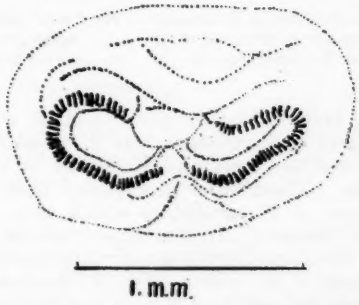


Fig. 4.—*E. scitula*, the symmetrical serrated rows seen on the last segment of caterpillar, Fig. 1, shown enlarged.

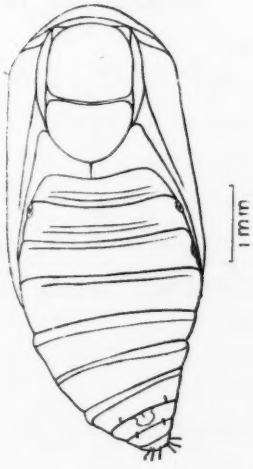


Fig. 5.—*E. scitula*, pupa, dorsal view, with short spines near the posterior end and long hook-like spines at the very end.

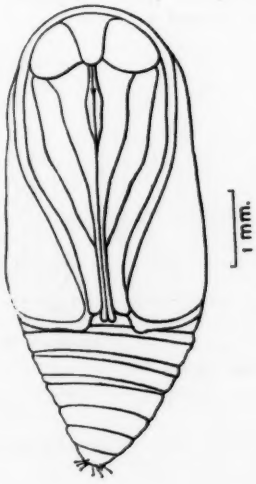


Fig. 6.—*E. scitula*, pupa, ventral view, showing the long hook-like spines.

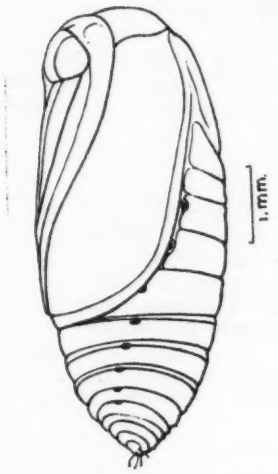


Fig. 7.—*E. scitula*, side-view showing the long row of spiracles and the terminal hook-like spines.

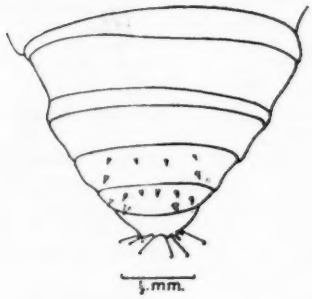


Fig. 8.—*E. scitula*, pupa, posterior end dorsal surface, enlarged to show short spines and the terminal long hook-like spines.

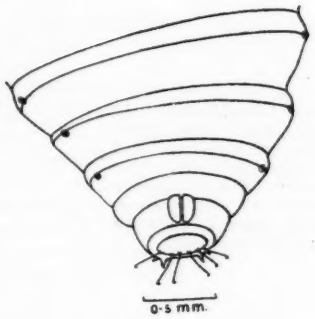


Fig. 9.—*E. scitula*, pupa, posterior end ventral surface, enlarged to show the terminal long hooks and the absence of short spines.

E. scitula both as a caterpillar and as a pupa comes nearer *E. coccidiphaga* than *E. amabilis*.The posterior end has short nail-like spines as well as long hooks. These differ in structure from those of the pupa of *E. coccidiphaga* which have been illustrated before.[2] All that now remains is to show the enlarged appearance of the posterior end of the pupa of *E. scitula*. Fig. 8 illustrates the dorsal posterior end and Fig. 9 the ventral end of the pupa further enlarged. The shorter spines are present only on the dorsal surface of the pupa as in Fig. 8 and absent on the ventral surface. The hooks of the longer spines are covered towards the ventral side, a feature which requires a careful comparison of Figs. 8 and 9.

With the help of these illustrations, the species *E. scitula* can be identified in the larval and pupal stages, even without any reference to those showing the pupae of *E. coccidiphaga* or of *E. ammabilis*.

References

1. S. Mahdihassan, Schemetterlingsraupen als Feinde der Lackschildause, *Natur u. Museum*, Frankfurt, Aug. 1929, Vol. 59, p. 394.

2. S. Mahdihassan, *Eublemma coccidiphaga*, J. Osmania Univ., **12**, 15 (1946).

CHAKSINE.[1] PART II.–PYROLYSIS OF CHAKSINE[2,3]

SALIMUZZAMAN SIDDIQUI, AHMAD KAMAL, GEORGE HAHN AND (IN PART) VISHWA NATH SHARMA

Central Laboratories, Pakistan Council of Scientific and Industrial Research, Karachi

(Received June 16, 1958)

In the course of studies in the constitution of chaksine, the principal alkaloid of *Cassia Absus Linn*, Siddiqui et al. observed theformation of a cyanide and an odorescent prod ct with a characteristic caraway-like smell un the thermal degradation of the salts of the bose. As a result of further studies in this direction, it has been noted by the present authors that the odorescent product obtained on heating chaksine iodide with Cu-bronze (Merck's) at 310°-320°C. in nitrogen atmosphere, gives on hydrolysis with 20% methanolic potassium hydroxide mainly phthalic acid along with a very small quantity of a light petroleum soiuble acid (m.p. 116°C.) which analyses for $C_{10}H_{12}O_2$. On the other hand, hydrolysis of chaksin iodide with copper filings or silver powder (40-80 mesh) at the same temperature range, yields an oily product, which on hydrolysis gives the light petroleum soluble acid m.p. 116°C. as the major constituent. This acid has been identified as *p*-isopropylbenzoic acid.

During the hydrolysiswith methanolic potassium hydroxide, ammoniaiwas evolved indicating the formation of the acd through a nitrile. The characteristic caraway-like smeli, on the other hand would indicate the presence of *p*-isopropylbenzaldehyde which on hydroiysis should furnish the corresponding acid through a Cannizaro reaction, but it was not possible to isolate the aldehyde from the pyrolysate.

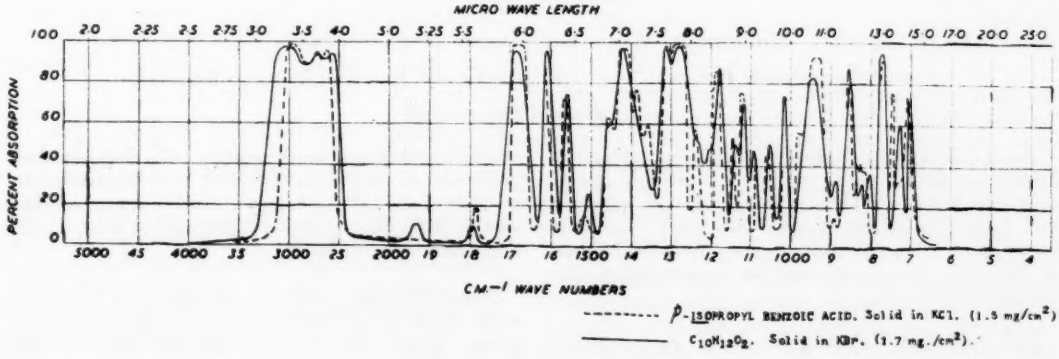
Experimental

All melting points are corrected.

Pyrolysis of Chaksine Iodide.—Chaksine iodide (1 g.) and copper bronze (1 g., Merck's) were thoroughly mixed and placed in a distillation flask (cap. 25 ml.) attached to a receiver tube. The mixture was heated to at 310-320°C. in a metal-bath under a slow current of nitrogen. The flask was removed after 25 minutes heating and the cooled contents extracted with ether repeatedly. The ethereal extracts were combined with the ethereal washings of the receiver flask, dried and the ether removed. The residual oil, deep brownish red in colour, had a characteristic caraway-like smell. Yield between 0.19-32 g. in different experiments.

The crystalline, ether-insoluble material collected from the receiver tube was identified as ammonium bicarbonate.

The oil (0.8 g.) was taken up in 20% methyl alcoholic potassium hydroxide (10 ml.) and refluxed on water-bath, ammonia being evolved in the process. After 10-12 hrs. heating methyl alcohol was removed in vacuum, the residue was taken up in water (5 ml.) and extracted with ether. The aqueous alkaline solution was acidified with dilute hydrochloric acid and repeatedly extracted with light petroleum (60-80°C.), and



Infrared absorption spectrum.

then with ether. Removal of light petroleum left an oily residue which on keeping gave a minute quantity of an acid which on several crystallisations from water melted at 116°C. and was identical with the acid described in the subsequent experiment.

Drying and removal of the ether gave material which crystallised from water (m.p. 196°C. dec.) with evolution of gas. Yield 0.12 g. It was identified as phthalic acid with which it gave no depression in melting point. On melting it gave the anhydride which was purified by sublimation. m.p. 130°C., undepressed with an authentic sample of phthalic anhydride. On heating with water gave phthalic acid m.p. 196°C.

Acid $C_{10}H_{12}O_2$.—Chaksine iodide (1.0 g.) thoroughly mixed with copper or silver filings (1 g. 40-80 mesh) was heated, in the manner described above, for 25 minutes. Yield of ether soluble red oil with characteristic caraway-like smell varied between 0.2-0.25 g. In one experiment 0.39 g. of the oil was obtained.

The ether soluble oil (0.85 g.) was taken up in 20% methyl alcoholic potassium hydroxide solution (10 ml.) and refluxed for 12 hrs. Methyl alcohol was removed in vacuum, the residue was acidified with dilute hydrochloric acid and repeatedly extracted with light petroleum (60-80°C.). Drying and removal of the solvent yielded an oil (0.18 g.) which crystallised on standing, m.p. 106°C. Crystallisation from water (twice) gave colourless prismatic needles, m.p. 116°C., (70 mg.). The compound is an acid and is soluble in all the usual organic solvents.

Calculated for $C_{10}H_{12}O_2$: C, 73.17; H, 7.32; O, 19.51; m.w., 164. Found: C, 73.39; H, 7.3; O, 19.38; m.w., 165.5 ; equivalent wt., 165.

On admixture with an authentic sample of *p*-isopropylbenzoic acid (m.p. 116°C.) the melting point was undepressed.

Acknowledgement

Micro-analysis was carried out by Dr. E. Bernhardt, Microanalytical Laboratorium, 22A, Mulheim, Ruhr, West Germany.

Our thanks are due to Dr. D.H. Whiffen and his associate Mr. H. Speddring of the Deptt. of Organic and Physical Chemistry, Birmingham University, for very kindly carrying out the two infra-red determinations, for comparison.

References

1. A. Kamal, and G. Hahn, J. Chem. Soc., 555 (1958).
2. S. Siddiqui, G. Hahn, V.N. Sharma and A. Kamal, Nature, **178**, 373 (1956).
3. S. Siddiqui, G. Hahn, V.N. Sharma and A. Kamal, Chem. and Ind. (London), 1525 (1956).

CHAKSINE. PART III

AHMAD KAMAL, MOHAMMAD AYUB BOKHARI, LOUIS FERNANDEZ AND GEORGE HAHN

Central Laboratories, Pakistan Council of Scientific and Industrial Research, Karachi

(Received June 16, 1958)

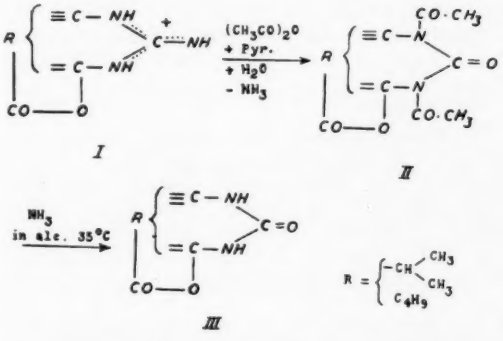
Acetylation of chaksine furnishes diacetyl-desiminochaksine (II), which accordingly yields desiminochaksine (III) on deacetylation, wherein the guanidine-imino group has been replaced by oxygen. The guanidine group first established in Part I is thus confirmed by conversion to the corresponding urea.

On hydrolysis of chaksine with an excess of aqueous barium hydroxide one molecule of ammonia is evolved within approximate 48 hrs. while it takes 200 hrs. more to develop another molecule of ammonia together with one molecule of carbon dioxide, leaving behind a water soluble and a difficultly water soluble amino acid, which are under investigation.

Even after one molecule of ammonia has been evolved the product of hydrolysis carries a carboxyl group (m.p. 123°C.). It follows, that besides the replacement of the guanidine-imino group by oxygen a carbonyl group containing, hydro-

lisable grouping of the molecule must have been split off simultaneously. This carbonyl group cannot stand adjacent to one of the imino groups of the guanidine, because it is well-known that such guanidine derivatives drop down considerably in their basicity. In comparison chaksine has still a $pK_a = 11\text{-}12$. It is therefore suggested that a lactone group is present accounting for the two oxygen atoms in chaksine. Consequently the two acetyl groups in diacetylchaksine must be attached at the two imino groups of the guanidine. On the other hand, the development of two molecules of ammonia and one molecule of carbondioxide out of a N,N'-disubstituted guanidine is only possible, if the oxy group of the lactone is combined with a carbon atom adjacent to one of the imino groups of the guanidine.

We therefore consider the structure (I) present in chaksine.



I II III

The C_{10}-moiety combined with guanidine makes it obvious that two isoprene molecules might have been involved in the phytosynthesis of chaksine, so much the more we have isolated *p*-isopropylbenzoic acid by pyrolysis of chaksine iodide with copper filings.[2] An isopropyl group must therefore be present and besides this there might further be a $-C(=)-$ group, because it is

$$\dot{C}H_3$$

not sure that the one molecule of acetic acid, received with the Kuhn-Roth oxidation stems from the isopropyl group.[3,4]

As it is possible to put up several monoterpene structures containing the guanidine moiety and explaining all the findings so far, we deliberately withhold to formulate any such suggestion until after more experimental evidence for one or the other is available.

Guha and Ray[5] described the acetylation of chaksine as allegedly resulting in the elimination of an aminoethanol chain, thus furnishing a "triacetyl compound" of the formula $C_{15}H_{20}O_5N_2$ m.p. 200°C. without any further confirmation.

According to the established guanidine structure of chaksine one acetyl group could be expected at the nitrogen atom carrying the nitro group in nitrochaksine sulphate. If so this must similarly result in a considerable drop in the basicity and perhaps a corresponding instability as observed with nitrochaksine sulphate. We therefore carried out the same acetylation, isolated the acetyl-product in 25% yield. m.p. 198-199°C. (dil. acetic acid) and found : C, 57·7 ; H, 7·5; O, 25·7; N, 9·2 ; which accounts as well for diacetyl-desiminochaksine, wherein the guanidine-imino group has been replaced by oxygen. $C_{15}H_{22}O_5N_2$ requires : C,57·9 ; H, 7·08 ; O, 25·7 ; N, 9.05. This seems so much the more obvious, because the oily acetylation product had to be heated for 10 hrs. with dilute acetic acid to become crystalline. In fact the deacetylation product with saturated alcoholic ammonia at room temperature (35°C.) furnished desiminochaksine, m.p. 253°C. (methanol, dil. acetic acid or water). Calculated for $C_{11}H_{18}O_3N_2$: C, 58·3 ; H, 7·95 ; O, 21·2 ; N, 12·3. Found : C, 58.66 ; H, 7·97 ; O, 21·57; N, 12.26. According to the urea structure, concentrated nitric acid forms at once the well crystallising nitrate which however on standing with water hydrolyses, furnishing back the starting desiminochaksine. On heating, the nitrate undergoes interesting changes still under investigation.

On carefully basifying the mother liquors of the acetylation, the diacetylchaksine m.p. 256°C. (dec.) could be isolated. It is unstable like nitrochaksine and easily undergoes hydrolysis. It therefore could not be purified reliably. Calculated for $C_{15}H_{23}O_4N_3$: C, 58.20 ; H, 7·5 ; O, 20.69 ; N, 13.58. Found : C, 57·31 ; H, 7.89 ; O, 19.62 ; N, 14.66. It is no more basic enough to form the guanidinium hydroxide.

The same drop in the basicity could be observed with nitrochaksine. Out of the basified mother liquors of the nitrochaksine preparation the free nitrochaksine similarly was found. Globulous clusters m.p. 166°C. (methanol). Calculated for $C_{11}H_{18}O_4N_4$: C, 48.8; H, 6.6; O, 23.7; N, 20.7. Found : C, 49.02 ; H, 6.85 ; O, 23,7 ; N, 20.4.

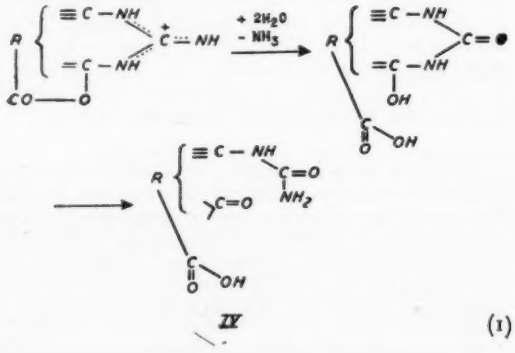
In confirmation of that sulphuric acid furnished nitrochaksine sulphate and on heating with dilute sulphuric acid the same hydrolysis to nitroureachaksine takes place; as observed in the case of nitrochaksine.

Most probably the more unstable nitro-

chaksinium hydroxide could also be isolated On repeated recrystallisation out of 16% alcohol the base m.p. 166°C. was more and more transferred into a base, difficultly soluble in alcohol and melting at 190°C. This base being amorphous and not so far recrystallisable could also be converted into nitrochaksine sulphate and subsequently into nitroureachaksine. Dissolved in chloroform and kept several days over dry sodium sulphate the base m.p. 166°C. could be recovered.

According to these findings we propose to use the name chaksine for the free guanidine, while the hydroxide and its salts should be considered chaksinium compounds.

On heating chaksine with aqueous barium hydroxide in the apparatus described in Part I[1] until one molecule of ammonia has evolved and removing the excess of barium hydroxide with carbon dioxide, a barium salt solution is obtained. This barium salt is highly hygroscopic and its acid, set free with ammonium sulphate and barium carbonate, is also hygroscopic and easily soluble in water. So far only a very small amount of that acid could be isolated. It melts at 123°C. and is under investigation. Since the necessarily present carbonyl group cannot be connected with an imino group of the guanidine, considering the strong basic character of chaksine, the remaining possibility is a lactone group where the oxy group must be connected with the carbon atom adjacent to one of the imino groups of the guanidine. The barium hydroxide hydrolysis can then be understood according to equation (1).



(1)

Besides hydrolysis of the imino group of the guanidine, furnishing the first molecule of ammonia, the lactone ring opens and an ureido acid IV is formed which might take 200 hrs. more for being hydrolysed further since it is well-known that urea itself requires extreme conditions (200°C. and a long time) to be hydrolysed by barium hydroxide.[6]

Experimental

Acetylation of Chaksine Sulphate. (a) *Diacetyldesiminochaksine* (II).—Chaksine sulphate (5 g.) in acetic anhydride (50 ml.) containing pyridine (0.5 ml.) was heated on water-bath (2 hrs.). Excess of acetic anhydride (ca. 45 ml.) was removed (vac.), and water (200 ml.) added. The contents were heated on water-bath (10 hrs.), cooled and the crystalline acetylated product filtered off. The diacetyldesiminochaksine was obtained as colourless prismatic needles. m.p. 194°C. (1.43 g ; 25.5%). On crystallisation from dilute acetic acid gave the product m.p. 198-199°C. Shows no optical rotation in glacial acetic acid or benzene.

Calculated for diacetyldesiminochaksine, $C_{15}H_{22}O_5N_2$ (I) : C, 58.02 ; H, 7.15 ; O, 25.8 ; N, 9.3. Found: C, 57.7; H, 7.5; O, 25.7; N, 9.2.

(b) *Diacetylchaksine.*—The aqueous filtrate was basified with liquor ammonia and the precipitate extracted with ethyl acetate, quickly dried over sodium sulphate and filtered. The solution slowly started depositing colourless prismatic needles. m.p. 256°C. (dec.), (0.25 g.). In some experiments a lesser yield was obtained. The product once it has crystallised out is insoluble in any of the bench solvents except ethyl alcohol and tetrahydrofuran in which it dissolves sparingly, a lacquer-like deposit being obtained on removal of the solvents. It could not therefore be purified properly.

Calculated for diacetylchaksine $C_{15}H_{23}O_4N_3$: C, 58.2 ; H, 7.5 ; O, 20.7 ; N, 12.58. Found : C, 57.3 ; H, 7.9 ; O, 19.6 ; N, 14.6.

Derivative of Diacetylchaksine. *Diacetylchaksine Nitrate :* from diacetylchaksine (0.1 g.) and dilute nitric acid. Crystallised from water. Prisms. m.p. 187°C. (dec.) $[\alpha]_D^{29.5}$ + 74.87 in water.

Calculated for diacetylchaksine nitrate : $C_{15}H_{24}O_7N_4$: C, 48.4 : H, 6.45 ; O, 30.11 ; N, 15.1. Found : C, 48.9 ; H, 6.81 ; O, 28.25 ; N, 15.5.

Removal of ethyl acetate from the mother-liquor after removal of diacetylchaksine gave an almost colourless weak amorphous base (A) (1.2 g.) soluble in ethyl acetate and methyl and

ethyl alcohols. It formed salts with mineral acids which hydrolyse on dilution with water.

Acetylation of Amorphous Base (A).—Amorphous base (A), (0.5 g.), taken up in acetic anhydride (5 ml.) containing a drop of pyridine was heated for 2 hrs. worked up as in the acetylation of chaksine sulphate. Colourless prisms m.p. 182°C., (0.16 g.), recrystallised from dilute acetic acid m.p. 198°C. undepressed with diacetyldesiminochaksine (II).

Acetylation of Chaksine Nitrate.—Chaksine nitrate (0.4 g.) in acetic anhydride (6 ml.) containing pyridine (2 drops) was refluxed on sand bath (5 minutes). Solution took place with evolution of oxides of nitrogen. Poured into water (80 ml.) and allowed to stand (24 hrs.). Residue crystallised from dilute acetic acid, prismatic needles, m.p. 197-198°C. (0.13 g.), undepressed on admixture with diacetyldesiminochaksine (II). Shows no optical rotation in acetic acid or benzene.

Acetylation of Base 256°C. (*Diacetylchaksine*).—Base 256°C. (0.1 g.) in acetic anhydride (1 ml.) containing pyridine (1 drop), heated on water bath (2 hrs.). Removed acetic anhydride in vacuum, added water (5 ml.), heated on water-bath (10 hrs.) and the colourless crystalline product collected. m.p. 187-188°C. Recrystallised from dilute acetic acid m.p. 198-199°C., undepressed with diacetyldesiminochaksine (II).

Desiminochaksine (III).—(a) *Deacetylation of diacetyldesiminochaksine with methyl alcoholic ammonia:* Diacetyldesiminochaksine (I) (4.7 g.) was taken up in methyl alcohol saturated with ammonia, (600 ml.) and kept at about 30°C. for 36 hrs. Removal of solvent from the clear solution gave diacetyldesiminochaksine as an oil which solidified on standing. Colourless prisms from hot water, dilute methyl alcohol or dilute acetic acid., m.p. 253°C., (1.8 g.). $[\alpha]_D^{34}$ + 72.76 in glacial acetic acid.

Calculated for desiminochaksine (III), $C_{11}H_{18}O_3N_2$: C, 58.3; H, 7.95; O, 21.2; N, 12.3. Found: C, 58.66; H, 7.97; O, 21.57; N, 12.26.

(b) *Deacetylation of diacetyldesiminochaksine with ethyl alcoholic HCl:* Diacetyldesiminochaksine (0.1 g.) taken in ethanolic HCl (1.8% ; 7 ml.) was refluxed for 10 hrs. Removed alcohol in vacuum and the oily residue extracted with hot water (10 ml.), cooled and filtered. Crystals after 7 days; m.p. 242°C. (10 mg.). Recrystallised from hot water ; m.p. 253°C. undepressed with desiminochaksine (III).

Found: C, 58.59; H, 8.02; O, 21.48; N, 12.16.

Acetylation of Desiminochaksine (III).—Desiminochaksine (III) (80 m.g.) taken up in acetic anhydride (3 ml.) containing a drop of pyridine was refluxed for 3 hrs. Removed acetic anhydride in vacuum and the residue crystallised from dilute acetic acid. Prismatic needles, (50 mg.), m.p. 198°C., undepressed on admixture with diacetyldesiminochaksine (II).

Desiminochaksine Nitrate. — Desiminochaksine (0.2 g.) was treated under ice-cooling, with nitric acid (d. 1.4 ; 5 drops). Prisms of the nitrate start coming out immediately. After standing (5 minutes) the crystallisate was filtered and pressed on a porous plate to remove the excess of nitric acid and kept in vacuum over sodium hydroxide. m.p. 133°C. (dec.) ; (0.15 g.).

Calculated for $C_{11}H_{18}O_3N_3 \cdot HNO_3$: N, 18.48. Found: N, 18.11.

Hydrolysis of Desiminochaksine Nitrate.—Desiminochaksine nitrate (0.1 g.) was covered with a little water and allowed to stand (3-4 hrs.), filtered and crystallised from dilute acetic acid (0.08 g.) m.p. 252°C. Mixed m.p. with desiminochaksine gave no depression.

Nitrochaksine Sulphate.—(*Isolation of free nitrochaksine*) : Chaksine nitrate (5 g.) was added in small portions and with rapid stirring to sulphuric acid (d. 1.84 ; 10 ml.), kept between −5°C. to 0°C., till clear solution was obtained. The contents, allowed to stand (45 minutes) at room temperature, were poured into ice (300 g.). Nitrochaksine sulphate, collected by suction, was washed with ice water m.p. 174-6°C. (dec.) ; (3.51 g.). The mother liquor on keeping in the cold for 24 hrs. gave a further amount (0.8 g.) of pure nitrochaksine sulphate, long prismatic needles. m.p. 180°C. (dec.). Total yield 4.3 g.

The aqueous mother liquor was basified with sodium carbonate carefully and the flocculent precipitate was extracted with chloroform. Removal of chloroform gave a colourless amorphous base. m.p. 166°C. (dec.) (1.12 g.) from methanol.

Calculated for $C_{11}H_{18}O_4N_4$, (270): C, 48.8; H, 6.6; O, 23.7; N, 20.7. Found: (1) C, 49.02; H, 6.85; O, 23.73; N, 20.47; (2) C, 48.87; H, 6.62; O, 23.90; N, 20.62.

Repeated crystallisation of the base 166°C. (dec.) from 96% ethyl alcohol changes the substance more and more into a difficultly soluble and unstable base melting at 190°C. (dec.) which

could not be properly recrystallised. Its solution in dry chloroform, on standing over sodium sulphate for a number of days, slowly gives back the original 166°C. (dec.) base.

The base which we believe to be the chaksinium hydroxide of the free chaksine base was characterised through its salts which were, as expected, identical with corresponding salts of the base m.p. 166°C. (dec.).

In view of the identity of the salts of the bases 190°C. (dec.) and 166°C. (dec.) the preparation of the salts of the former alone are being reported.

Sulphate of Base 190°C. (dec.) (*Nitrochaksine Sulphate*) : The base (0.2 g.) was dissolved in portions in conc. sulphuric acid (d. 1.84 ; 0.5 ml.), maintained at −5°C., with constant stirring and rubbing, (1.5 hr.). Poured into ice (15 g.) and the product collected was washed with ice-cold water ; m.p. 162-164°C. (dec.) ; (0.16 g.). mixed m.p. with nitrochaksine sulphate 172°C. (dec.).

Since the sulphate decomposes it was not purified by crystallisation from water but was hydrolysed to nitroureachaksine.

Nitrochaksine Oxalate.—Base 190°C. (dec.) (0.2 g.) in dioxane (2 ml.) added to dioxane solution of oxalic acid. Golubles of nitrochaksine oxalate; m.p. 191°C. (dec.) (0.2 g.)

Calculated for nitrochaksine oxalate $C_{13}H_{20}O_2N_4$: C,43.33 ; H,5.55 ; O,35.55 ; N,15.55. Found : C,43.51 ; H,5.87 ; O,35.72 ; N,14.97.

Nitrochaksine Chloride.—Base 190°C. (dec.), (0.2 g.), dissolved by warming in concentrated hydrochloric acid (1 ml.). Cooled and poured into water. Colours at 200°C. m.p. 215°C. (dec.).

Calculated for nitrochaksine chloride $C_{11}H_{19}O_4N_4Cl$: C, 43.05 ; H, 6.25; O, 20.88; N, 18.27 ; Cl, 11.5. Found : C, 42.97 ; H, 6.59 ; O, 21.10 ; N, 18.39 ; Cl, 11.13.

Solution of the chloride in water on basification with ammonia gave the original base m.p. 190°C. (dec.), undepressed on admixture with the starting material.

Hydrolysis of Sulphate of Base 190°C. (dec.).—The sulphate (0.43 g.) taken in water (43 ml.) was heated on water bath. After ½ hr. *p*H was 4.8 which came down to 2.5 after 8-10 hrs. After 36 hrs. heating was stopped and the crystalline product collected (0.19 g.) ; m.p. 200°C. (dec.), undepressed with nitroureachaksine.[1]

Direct Hydrolysis of the Base 190°C. (dec.).- The base (0.3 g.) suspended in water (30 ml.) containing 2 drops of 10% sulphuric acid was heated on water bath. The solution which had *p*H 1 at the start became clear (½ hr.) and had *p*H 3-4 after 13½ hrs. After 36 hrs. (*p*H 2.5) the reaction was stopped and the colourless crystalline material collected. (0.13 g., 41.9%), m.p. 199-200°C. (dec.) undepressed with nitroureachaksine.[1]

Reaction of Nitrochaksine Sulphate with Sodium Carbonate.—Nitrochaksine sulphate (1.06 g.) was taken up in a solution of sodium carbonate (5.3 g. in 35 ml. water) by warming for ½ hr. at 50-70°C. After standing for 3 hrs. the solution was partly acidified with HCl, flocculent matter (0.1 g.) filtered off and the mother liquor further acidified with HCl (*p*H 2). A further quantity of a white flocculent precipitate (0.3 g.) was filtered off; m.p. 160-170°C. (frothing). This material is soluble in ethyl and methyl alcohols. With water gets oily on warming and forms a barium salt. Partly soluble in acetone, the acetone insoluble portion being soluble in acetic acid and hot methyl alcohol, and forms an insoluble barium salt. The two fractions are under investigation.

The filtrate now on being neutralised with sodium carbonate (*p*H 8-9) gave a white flocculent precipitate which crystallised as globules from dioxane and ethyl alcohol (40 mg.). Turns brownish red at 160°C. melting at 166°C. (dec.). Mixed m.p. with the base 166°C. (dec.) (obtained in the reaction of chaksine nitrate and sulphuric acid) undepressed. It is most probably free nitrochaksine.

Acknowledgement

Micro-analysis was carried out by Dr. A. Bernhardt, Microanalytical Laboratorium, 22A, Mulheim, Ruhr, West Germany.

References

1. Ahmad Kamal and George Hahn, *Chaksine Part I*, J. Chem. Soc., 555 (1958).
2. S. Siddiqui, George Hahn, A. Kamal and V.N. Sharma, J. Soc. Chem. Ind. (London), 1525 (1956).
3. Richard Kuh and Hubert Rath, Ber., **66**, 1274 (1933).n
4. W.F. Barthel and F.W. LaForge, Ind. Eng. Chem. (Anal. Ed.), **16**, 434 (1944).
5. S.K. Guha and J.N. Ray, J. Indian Chem. Soc. **33** (4), 225.
6. E.A. Werner, J. Chem. Soc., **113**, 84 (1918)

NON-TECHNICAL NOTES

COTTONSEED OIL BASED PAINTS AND OTHER COATING COMPOSITIONS

Drying oils form an essential ingredient of paints, varnishes, and other coating compositions. The resources of drying oils like linseed and tung oils being limited in our country, research work was started at the Central Laboratories on the improvement of the drying property of cotton seed oil, a semi-drying oil abundantly available in the country.

Pakistan produces about 600,000 t ns of cottonseed annually, the area under cultivation in 1953-54 being 2,928,000 acres. Of this about 50,000 can be deducted to allow for requirements of seeds and wastage, leaving a balance of 5,50,000 tons. Calculating on the basis of an average yield of 12 % of refined oil, approximately 69,000 tons of cottonseed oil can be produced in the country. This figure will be still higher when the Government target of increasing the cotton production by another 40% by the end of 1957, is achieved (see also appendix A & B).

Cottonseed oil has so far been used in the paint industry only to a limited extent as an adulterant for the usual drying oils. It can be combined to a maximum of 20-25% with linseed oil and about 30-35% with tung oil for the making of oleo-resinous varnishes, without impairing the quality of the films produced. A larger proportion of cottonseed oil produces a tacky film.

As a result of researches carried out at the Central Laboratories, it is now possible to eliminate the natural drying oi's and to use cottonseed oil alone for the preparation of oleo-resinous varnishes. In their performance, these varnishes compare favourably with similar compositions made from linseed oil. These coating compositions can be used :—

1. as a varnish.
2. for the manufacture of general-purpose quick drying paints by the incorporation of appropriate pigments.
3. for emulsion paints (oil bound washable distempers).
4. for water proofing packing paper.

Different qualities of the varnishes can be prepared to suit various requirements and may be classified as follows :—

Class 'A': wherein the acidity of rosin is decreased by lime alone or by a mixture of lime and zinc-oxide.

Class 'B': wherein the acidity of rosin is reduced partially by lime and zinc-oxide and partially by esterification with glycerol.

Class 'C': wherein the acidity of rosin is reduced merely by esterification with glycerol.

The greater the quantity of glycerol used for neutralisation of the rosin, the better is the quality of the varnish obtained. Class 'C' is therefore superior to Class 'B', which in turn is superior to Class 'A'. The quality is further dependent on the oil length of the varnish. The greater the oil length the greater is the flexibility and water resistance. (Oil length signifies quantity of oil in gallons per 100 lbs. of rosin.)

Process

The process consists mainly of cooking the oil with rosin and metallic salts under controlled conditions of temperature and time.

Equipment

No special equipment is required. Ordinary open varnish-cooking kettles usually installed in paint and varnish factories in this country can serve the purpose. These kettles can be fabricated locally.

The adoption of the process developed at the Central Laboratories should eliminate the use of linseed oil for the manufacture of :—

1. Oleo-resinous varnishes and paints (oil length '1' to '18' gallon varnish).
2. Oil bound washable distempers.

Furthermore with current prices of the oils, cottonseed oil based coatings would be about 20% cheaper than the corresponding linseed oil coatings.

All the materials required for the manufacture of these coatings based on cottonseed oil are available in the country with the exception of a few chemicals.

The total capital investment for a unit with a production capacity of 36,000 gallons per annum is Rs. 100,000. The average cost of the cottonseed based coating composition is Rs. 7 per gallon. In comparison the average cost of a similar composition based on linseed oil would be about Rs. 9/4/- per gallon.

Pre-Construction Cost Estimation

Production Capacity :—

Class 'A' 12,000 gallons per annum.
Class 'B' 12,000 gallons per annum.
Class 'C' 12,000 gallons per annum.

I. Capital Expenditure

		Rs.	Rs.
A.	Equipment ..	15,000	
B.	Building ..	27,000	
	Total capital expenditure ..		42,000

II. Running Expenditure

1.	Direct Expenses per annum		
	Raw materials ..	1,51,556	
	Direct wages ..	13,020	
	Containers ..	45,000	
	Fuel ..	2,500	
	Contingencies ..	5,000	
	Depreciation @ 10%	4,200	
			2,21,276
	Establishment ..	1,5600	
	Promotion of the project @ 2½% on Rs. 1,00,000/-	2,500	
			18,100
2.	Selling expenses		4,200
3.	Interest on capital @ 4% on Rs. 1,00,000/-		4,000
4.	Insurance @ 2½ % on Rs. 2,00,000/-		5,000
	Total cost of production for 36,000 gallons ..		2,52,576
	Average cost of production of 1 gallon.		Rs. 7

Appendix A

1. Quantity of cottonseed available in Pakistan .. 6,30,000 tons
2. Quantity of cottonseed used as cattle food .. 1,00,000 tons
3. Quantity of cottonseed oil actually produced .. 50,000-60,000 tons

(These figures have been obtained from Seed Merchants' Association).

Installed capacity of the plants for hydrogenation is 22,000 tons. Actual production is about 14000-15000 tons.

(Information from D.G.S.&D., Development Wing).

Appendix B

LOCAL PRODUCTION OF LINSEED OIL

Year	Area under cultivation (acres)	Linseed production (tons)	Linseed oil production (tons)
1953-54 ..	74,000	12,000	3,000
1954-55 ..	76,000	14,000	3,500

IMPORT OF LINSEED OIL

Period	Quantity Galls.	Quantity Tons	Value Rs.
July '50-June '51 ..	83,024	297	3,43,448
July '53-June '54 ..	98,944	354	4,17,040
July '55-Dec. '55 ..	100,302	359	7,62,360

N.B.—Exact figures for the local production of linseed oil are not known. The quantity shown in the table indicates the probable production of the oil, based on the information of available seeds.

SHARK LIVER OIL AND VITAMIN A CONCENTRATES

Fish fauna of the Arabian sea is a natural resource of great significance in the economy of Pakistan. The long West Pakistan coast offers exceptionally good opportunities for developing fisheries on a commercial scale. Along with the edible fishes a considerable number of sharks are landed throughout the year mostly at four or five coastal fishing villages along the Makran coast and around Karachi.

Shark liver oil is known to be a rich source of vitamin A. There are several other marine fishes that contain vitamin A in their livers. To name a few, cod, hallibut, sword-fish etc., are exploited for the extraction of liver oil which is sold in the world markets after the necessary refining and standardization. Pakistan imports large quantities of such liver oils for its vitamin A and D content along with other preparations containing synthetic vitamin A, and the country thus incurs a considerable amount of foreign exchange expenditure.

In view of the facts stated above, investigations were started over a year ago in the Central Laboratories of the Pakistan Council of Scientific and Industrial Research to study the possibilities of developing the shark liver oil industry in the country. As a result of these studies a process has been evolved for the simultaneous production of concentrates of vitamin A with a potency of around 1 million U.S.P. units per gram, and a deodorized, decolorized oil with a standardized vitamin A content of around 7000 U.S.P. units per gram. After completion of laboratory experiments the economics and optimum working conditions of the process have been thoroughly studied on a pilot plant.

The total capital investment for a unit with an annual production capacity as given below is estimated at approximately Rs. 3,30,000/-.

1. Refined shark liver oil, 7000 U.S.P. units per gram, approximately 16,800 lbs.
2. Vitamin A concentrate, 10,00,000 U.S.P. units per gram, approximately 34 lbs.
3. Emulsion, 2,400 U.S.P. unit per gram, approximately 40,000 lbs.

Total vitamin A content of the above products will be approximately 73 thousand million U.S.P. units, giving an average cost of Rs.-/5/6 for 1,00,000 U.S.P. units of vitamin A.

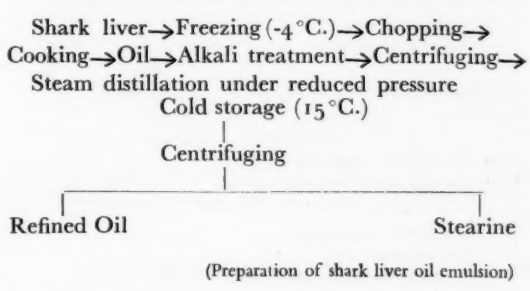
The retail market price of imported cod liver oil, *e.g.*, Seven Seas Cod Liver Oil, is Rs. 1/12/- for 8 oz. containing 1,60,000 U.S.P. units of vitamin A and 20,000 U.S.P. units of vitamin D.

If it is desired to produce an oil with both vitamins A and D, the latter may be added in the form of calciferol, at an additional cost of Rs. -/5/- for 20,000 U.S.P. units of vitamin D, giving the cost of the finished product comparable to Seven Seas Cod Liver Oil as below :—

1,60,000 U.S.P. units of vitamin A in shark liver oil	Rs. -/9/-
Additional cost of vitamin D, 20,000 U.S.P. units	Rs. -/5/-
	Rs. -/14/-

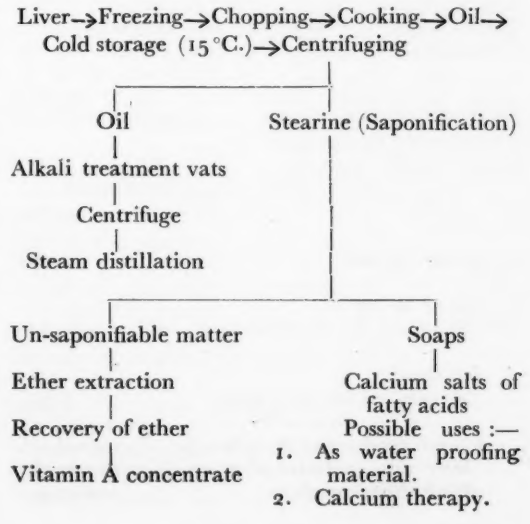
Process

The process follows the flow sheet given below :



(Preparation of shark liver oil emulsion)

A part of the oil can be used for the preparation of vitamin A concentrate as given in the flow sheet below :—



Equipment and Raw Materials

The equipment required consists of the following :—

Refrigeration units for cold storage, liver chopper, steam jacketted cooking pans, Sharples centrifuge, alkaline treatment vats, steam distillation apparatus, crude oil electric generator, overhead tanks, boiler, capsulating machine, spectrophotometer and other analytical equipment for the testing and standardization laboratory, homogenizer, Soxhlet type continuous extractor, and equipment for processing the by-products.

The raw-materials are shark liver and chemicals.

Summary of Pre-Construction Cost Estimation

(to process ¼ ton shark liver oil per day)

		Rs.	Rs.
1. *Capital Expenditure*			
Equipment	..	1,80,000	
Building	..	92,000	
Total capital expenditure	..		2,72,000
Supervisory staff and workers			54,420
2. *Running Expenditure (per annum)*			
Raw materials	..	63,645	
Direct wages	..	31,800	
Containers	..	50,000	
Power	..	10,500	
Contingencies	..	5,000	
Maintenance	..	9,600	
Depreciation @ 10%	..	27,212	
			1,97,757
Establishment	..	22,620	
Promotion of the project @ 2½% on Rs. 3,30,000/-	..	8,250	
			30,870
3. *Selling expenses*	..		7,200
4. *Interest on capital* @ 4% on Rs. 3,30,000/-			13,200
5. *Insurance* @ 2½% on Rs. 3,35,800/-			8,395
Cost of production of 33,600 lbs. of crude shark liver oil, inclusive of processing expenses for the finished products	..		2,57,442

SUBSTITUTE FOR BIDI LEAVES

The importance of bidi leaves (commonly called Tendu leaves and imported from India) for the local bidi industry is well known. On account of its high cost and import restrictions, a search for alternative leaves of Pakistani origin assumed great urgency immediately after independence. As a result of investigations carried out in the Council's Central and East Regional Laboratories, it has been possible to evolve a process for utilising the Kumbi leaves, found extensively in the forest area of Bahawal, Rajendrapur, Srepur etc. and obtainable at a nominal royalty of Rs. -/1/- per maund to the Forest Department.

The leaves as such, after drying, cannot be utilised as bidi wrappers, because of the disagreeable smell on burning and a number of other drawbacks *e.g.*, the difficulty of obtaining a continuous burning, wrinkle formation, and greenish colour persisting even after drying. These difficulties with the Kumbi, which is otherwise a tough leaf, have been overcome satisfactorily by a simple chemical and physical processing which can be carried out on a cottage industry scale.

Considerable quantities of bidies were made in the Laboratories with processed Kumbi leaves by a skilled Bidi maker using the same tobacco as used in bidies made out of imported bidi leaves from India. These bidies were given an extensive use as trial during the PIIF exhibition to a large number of visitors, and it can be said on the basis of reports received from the users, that the bidies made with treated Kumbi leaves are just as satisfactory as those made from Indian bidi leaves. The process can be carried out on a cottage industry scale with a total capital investment of about Rs. 1000 and on the basis of cottage production the cost of one seer is estimated to be about Rs. -/5/8.

Process

The leaves are boiled within 72 hours of plucking or after drying. The freshly collected leaves are dipped into boiling water for half an hour to forty minutes. These are then washed with fresh cold water several times and then processed with a chemical solution as below. The hot treated fresh leaves assume a light yellow colour somewhat similar to bidi leaves.

The leaves are next steeped overnight in a chemical solution (1%) in wooden vats and then taken out and air dried. When the moisture

content falls sufficiently, the leaves are rolled between heated rollers in a simple hand-operated machine designed for the purpose in the Council's Laboratories. As they come out of the roller, they are suitable for packing and marketing.

Equipment and Raw Materials

Equipment required for this industry consists of a series of wooden vats, a few aluminium receptacles for boiling, coal or wood fired heating ovens, and a retting machine which can be fabricated locally according to specifications.

The raw-materials required for this industry are Kumbi leaves which are easily available in East Pakistan and a small amount of chemicals.

Cost of Production

	Rs.	a.	p.
Capital expenses for equipment	570	0	0
Cost of raw materials for 20 seers of finished leaves	2	7	1
Repairs, maintenance, unskilled labour & miscellaneous (water lighting, etc.)	3	10	8
Depreciation, rent and taxes	1	0	6
Cost of production of 20 seers of finished leave	7	2	0
Cost of production per seer	0	5	8

PREPARATION OF MIXED BILE SALTS AND ACIDS FROM BILE

Mixed bile acids and salts are used in a number of pharmaceutical preparations and media for the cultivation of organisms. The pharmacopial preparations include desiccated bile, bile extract, acidium dehydrocolicum and its sodium salt. A large number of properietary drugs (shown in the appendix) are sold in the market in which the principal constituents are mixed bile salts and bile acids. A fairly large amount of bile salts also find use in the preparation of media for the cultivation of certain types of organisms in bacteriological laboratories.

Substantial amounts of bile are available in the slaughter houses in different parts of the country.

Approximately 13,000 lbs. of ox bile annually is available in one of the slaughter houses in Lahore alone. It is estimated that almost all the requirements of the country should be met from sources available within the country. Other products which can be made from mixed bile salts and bile acids are (1) mixed bile acids and salts (2) cholic acid (3) desoxy-cholic acid (4) choleic acid. These also find use in therapeutics preparations and in special media.

The most easily marketable product and the one which has a large demand is mixed bile salts and acids. It can be readily prepared from suitably treated bile. This commercial product of mixed bile acids and salts does not need the setting up of an elaborate factory but could be taken up by pharmaceutical concerns in a relatively small shed of 15′×15′.

The total capital investment in a unit with a production capacity of 12,000 lbs. per annum is estimated at Rs. 6,560.

It has not been possible to ascertain the prevailing market prices of these materials. But on the basis of finished proprietary drugs marketed in the form of tablets etc., it is estimated that the cost of production in the proposed unit compared to the prevailing cost will be of the order of 1 to 20.

Raw Materials and Equipment

Raw materials: Bile, commercial sodium hydroxide, and commercial sulphuric acid.

Equipment: Kettles made of copper, 100 lbs. capacity; 4 condensers spiral type 3′×1′ and 1′ diameter with water jacket 4′×2′ made of iron sheet of 24 gauge and suitable angle iron stand ; one storing drum ; one mixing tank ; 12 coarse wooden trays ; and one pestle and mortar.

Pre-Construction Cost Estimation

	Rs.	Rs.
Capital Expenditure :		
Cost of equipment, erection of a stove and shed ..		2,375
Running Expenditure per mensem :		
Cost of 1000 lbs. of bile	250	
Cartage ..	60	
Cost of chemicals ..	190	
Fuel ..	400	
Establishment ..	495	
		1,395

Yield of mixed bile acids from 1000 lbs. of liquid bile	44 lbs.
Cost of one pound of mixed bile acids	Rs. 32

Appendix

Bilisalin : Sold as tablets each containing bile salts 4 gr. and hepatic substance 2 gr.

Bilron : Sold as capsules each containing 5 gr. iron bile salts.

Desibyl Capsules : Each containing desicoated whole bile 5 gr.

Glanfel Tablets : Enteric-coated tablets 1 or 3 gr. consisting chiefly of sodium glycocholate and sodium taurocholate in proportion existing in the fresh bile. This is nothing but mixed bile acids.

Hydro-Bilein : Tablets each containing dried purified ox bile 2 gr. and dehydrocholic acid 2 gr. which is an oxidation product of cholic acid.

Felamin : Hexamine (0.225 g.) with cholic acid (0.075 g.).

Veracolate, Bicolate and Biledase : Contains sodium taurocholate and glycocholate with cascara, phenophthalein and capsicum.

Taxol Pancrobilin, Desicol and Jubol : All these preparations contain mixed bile salts.

INSULATING TAPES

A dielectric or insulating material may be characterised roughly as having an electric conductivity of less than 10^{-6} mhos/cm. Materials with conductivities in the range of 10^{-6}, 10^{-3} mhos/cm are semi/conductors. Usually, mechanical, chemical, thermal and cost considerations determine the choice of an insulating material. The cheapest and the simplest form of insulation is the insulating tape made of cloth or paper and coated with an insulating material. Even this type of insulating material is imported in Pakistan. It is estimated that Pakistan consumes annually about 3 lac spools of 1/2″ width and 50 yards length. A spool of these measurements imported from abroad costs about Rs. 2/- to Rs. 2/8/-

As a result of investigations carried out at the Central Laboratories of the Council of Scientific and Industrial Research, Karachi, it has been possible to evolve a process for the manufacture of insulating tape from exclusively indigenous raw materials. The tape is about 100% above the British Standards Specifications (B.S.S.).

The total capital investment for a unit of production for manufacturing 300,000 spools (1/2″×50 yards) per annum is estimated at approximately Rs. 2,14,000. The average cost of a spool in a unit of this size works out to Rs. 1/7/-, inclusive of overhead expenses, as against the present market price of a spool *viz.* Rs. 2/- to Rs. 2/8/-.

Process

The process involves the following unit operations : mixing the various ingredients, coating of the tape, drying of the tape at controlled temperature, and winding of the tape and cutting into spools of required sizes.

Equipment and Raw Materials

The equipment required for the process consists of : jacketed vessel with mixing and temperature control device, bath for coating purpose, drying chambers, and winding and cutting machine.

The raw materials required are : insulating medium, vehicle, filler and cloth.

All these materials are available in the country.

Summary of Pre-Construction Cost Estimation

(Production Capacity—3,00,000 spools per annum or 100 spools per day)

		Rs.	Rs.
I. Capital Expenditure			
Equipment	..	70,000	
Building	..	43,000	
Total capital expenditure	..		1,13,000
II. Running Expenditure (Per annum)			
1. Direct Expenses			
Raw materials	..	3,41,100	
Direct wages	..	13,320	

		Rs.	Rs.
	Containers ..	3,900	
	Power ..	10,000	
	Contingencies ..	6,000	
	Depreciation @ 10%	11,300	
	..		3,85,620
2.	Indirect expenses		
	Establishment ..	12,840	
	Promotion of the project @ 2 % on Rs. 2,14,000	5,350	
			18,190
3.	Selling Expenses ..		5,040
4.	Interest on capital @ 4% on Rs. 2,14,000/-		8,560
5.	Insurance @ 2½% on Rs. 4,54,100/- ..		11,353
	Cost of production of 3 lacs spools of tape ..		4,28,763

Average cost Rs. 1/7/- per spool.

PETROL GAS PLANT

A regular supply of gas is one of the essential requirements of a laboratory. The gas is usually obtained from the city gas supply, or generated in large plants of Mansfield type, installed at site. So far there is no city gas available anywhere in Pakistan and although with the discovery of the Sui Gas it is expected that gas will be available for domestic consumption in some of the cities, an independent gas plant will continue to be a necessary item of laboratory equipment. While some of the larger institutions have installed Mansfield type gas plant, most of the smaller industrial laboratories, have been experiencing considerable difficulty in this respect. During the last few years imported gas plants of the Aerogen type have been obtained at considerable cost from abroad, but on account of the fact that even a small size gas plant sufficient for 60 burners costs about Rs. 8000, a good part of which amount represents foreign exchange, a large number of smaller institutions have practically no arrangements for the supply of gas. In order to meet this requirement the Chemical Engineering Section of the Central Laboratories of the Council of Scientific and Industrial Research took up the design and fabrication of a portable, low-cost gas plant. The gas plant, of which many units have already been fabricated in the workshop of the Laboratories, has the following general specifications :—

1. 500 cu. ft. gas/gallon petrol.
2. 300 B.T.U-/cu. ft. of gas.
3. 300 cu. ft. of gas/hr. for 60 burners.
4. Overall dimensions : 4′-8″×2′-4″×5′-2″.
5. Power required : 185 watts.

The cost of the plant has been worked out to be about Rs. 1300. All parts of the plant can be fabricated except a high speed motor which will have to be imported. On the basis of comparative performance under similar conditions, the gas plant fabricated in the Council has been found to possess many advantages over similar imported gas plants. Some of the special features of the plant are, that it gives a longer flame, takes less time to generate the same quantity of gas, maintenance is easier, wear and tear is much less and the gas plant is more compact, requiring much less floor space compared to imported gas plants of the same capacity.

The fabrication of the gas plant can be taken up in any good workshop at an estimated cost, as indicated below :—

Summary of Pre-Construction Cost Estimation

		Rs.	Rs.
I. Direct Charges			
A.	M.S. sheets and other materials ..	250	
	Fabrication of carburetting unit ..	150	
	Driving unit and accessories ..	400	
B.	*Labour* ..	300	
			1,100
II. Indirect Charges			
1.	Overhead charges ..	200	
2.	Electric charges ..	50	
3.	Depreciation on machinery	50	
4.	Contingencies @ 5% on labour and material ..	50	
5.	Unforseen expenses ..	75	
			425
Cost of production of Petrol Gas Plant ..			1,525

NOTES & NEWS

Pyrethrum for Resistant Varieties of Insects

It is well known that in regions where regular and large scale spraying with DDT and other synthetic insecticides is practised, mosquitoes, flies and bugs are becoming resistant to them. This poses a serious problem, an answer to which is suggested by the fact that no insect ever managed to resist pyrethrum, the oldest insecticide on record and known to have been used by the Chinese some 2000 years ago. Pyrethrum comes from a daisy-like flower grown in Kenya, and it has been established that its insecticidal properties are due to two active constituents, pyrethrins I and II. Direct extraction of pyrethrum flowers with mineral oils gives extracts with a maximum pyrethrin content of only about 1%. Concentrates with higher pyrethrum contents are generally prepared by exhaustive extraction of the flowers with low boiling organic solvents and removal of the inert material from the total extractive by chilling in a freezing mixture.

S. Siddiqui and A.V. Subbaratnam [Indian Patent No. 35338 (August 8, 1946)] have developed a cheaper and better method for the preparation of concentrated extracts. The basis of this method is to carry out extraction of the flowers with a solvent mixture consisting of kerosene, alcohol and acetone. Petrol, white spirit, or any other mineral oil may be used as a substitute for kerosene and Ansol M and methyl acetone for alcohol and acetone, respectively. On removal of alcohol and acetone under reduced pressure, a concentrated solution with a pyrethrin content of 8 to 10% is obtained.

The concentrated extracts are commonly used in suitable dilutions as sprays for household, horticultural or livestock purposes. The extracts, however, lose their insecticidal activity on storage, because of oxidation and decomposition of pyrethrins through the effect of heat, light and air. To stabilise the extract, an anti-oxidant such as bhilawanol, sulphonated bhilawanol, or sulphonated bhilawan shell liquid, prepared from Bhilawan (marking nut), can be added as shown by S. Siddiqui, A.V. Subbaratnam and V. Sharma [Indian Patent No. 35567 (September 3, 1946)]. Other anti-oxidants which can be used are α-naphthylamine, hydroquinone, thymol, lecithin and pine oils.

The extracts prepared by this process are particularly well-suited to the preparation of fine quality semi-vanishing mosquito-repellent creams, in so far as smaller quantities of kerosene extracts are required to give the desired pyrethrin content and the creams are therefore more stable towards storage or transport in hot climates.

Safflower Oil for Troubled Arteries

Hardening of arteries, which is an ever present danger after the age of forty, increases the likelihood of heart attacks and is the result of excessive presence of cholesterol in the blood. It has now been more or less confirmed that the quantity of cholesterol in the blood can be reduced by the use of oils containing specific unsaturated fatty acids like linoleic acid.

Safflower oil contains 75-80% of linoleic acid, and has been lately marketed in the U.S.A. by various firms for use as a salad or cooking oil and also in the form of emulsions to be taken medicinally. This oil is obtained from a thistle-like plant, "*Carthamus Tinctorious*," long grown in India, Egypt, Spain, Italy and S. Russia for the red dye (carthanium) obtained from its flowers and also for its edible oil. Its cultivation in the U.S.A. is of recent origin, and special oil bearing varieties have been developed, which are reported to contain oil to the extent of 38% of the whole seed, as against 25-28% reported for seeds cultivated in other countries. Work in this direction has also been carried out with considerable success in Pakistan since 1951 by Messrs S.L.D. Farms, Mirpurkhas (Sind), the oil content of the various varieties cultivated being found to be nearly 35%.

Power from the Fusion of Hydrogen Nuclei

Towards the end of January, British scientists of the Atomic Energy Authority announced their success in carrying out the hydrogen fusion reaction under controllable conditions. This is the kind of reaction that takes place inside the sun and other stars, and supplies the tremendous quantities of energy that have kept these bodies hot for several thousand million years.

The successful experiments were done with the Zero-Energy Thermonuclear Assembly, called the "ZETA" reactor, in which heavy hydrogen in a doughnut-shaped toroidal tube is heated by a

large transient electric current (upto 200,000 amperes). The hot hydrogen is kept in the middle of the tube by the application of a toroidal magnetic field of several hundred Gauss. This device fences in the ionized gas and provides a thermally insulated zone of high temperature (at nearly 5 million degrees centigrade), in which the fusion of hydrogen atoms to build up heavier nuclei takes place with the liberation of several M.E.V. of energy per molecule. The experiments have so far been of a pilot nature and work is in hand on another ZETA that will produce temperatures of over ten million degrees centigrade. Physicists are confident that the further development of these experiments will, within a period of 30 years or so, provide mankind with an unlimited power from the hydrogen contained in the water of the oceans of the world.

Tenth Pakistan Science Conference

The 10th Pakistan Science Conference was held in Lahore this year from the 10th to the 15th March. The Conference was inaugurated by Dr. Nazir Ahmed, Chairman of the Atomic Energy Commission, after which Dr. M.O. Ghani, Agricultural Development Commissioner, East Pakistan, delivered a thought-provoking presidential address on the "Development of Science in Pakistan," in which he urged the necessity of co-ordinated development of science in the schools, universities, and research and technical institutions. He proposed the setting up of a National Science Development Commission to take up the problem as an organic whole.

Delegates from several foreign countries were present at the inaugural session and several others arrived later during the course of the Conference. Among the addresses of the sectional Presidents, a refreshing feature was that the President of the Engineering Section presented an outline of a possible hydro-electric project in the Kaghan Valley, which could be expected to contribute 160 megawatts of electrical power at a very low cost.

An outstanding feature of the Science Conference was the large number of well-attended scientific symposia on topics including the scientific and technological development of Pakistan. Notable among these were : Sui gas and its industrial possibilities ; *Rauwolfia serpentina* and some of the most recent trends in Rauwolfia therapy ; artificial satellites and space travel ; processing of low-grade iron ore ; and exploration and development of mineral resources of Pakistan.

Foreign delegates spoke in several of these symposia. Among the general symposia, those on the "Role of Scientists in Modern Society" and "Efficient use of Scientists" deserve mention. Lively interest was particularly evinced in the former. Diverse views were expressed by the speakers, although they generally agreed that scientists should concentrate their activities on their specialized fields, where they could utilize their talents to the greatest benefit of society.

Scientific and Industrial Research in Australia

Sir Ian Clunies Ross, Chairman, Commonwealth Scientific and Industrial Research Organization, Australia, delivered a lecture on "Some Aspects of Scientific and Industrial Research in Australia" at a function arranged by the Pakistan Association of Scientific Professions at the Y.M.C.A. Hall, Karachi, on the 3rd February, 1958. Dr. Salimuzzaman Siddiqui, Director, Scientific and Industrial Research, presided.

Sir Ian, while appreciating the Australian Government's approach towards scientific research in providing funds for it, said that from the experience of his organization he could say that for every pound invested in research at least ten pounds come back to the country and community. He emphasized that such research cannot always be directed but must often be given a free hand. This freedom, he said, contributed greatly to whatever success the Organization had achieved.

Sir Ian mentioned some instances where the Organization had been of service to the people. The first case he referred to was how the Organization by introducing *Cactoblactus* parasite in 1926 had been able to wipe out the cactus infestation which had affected 60 millions acres of land.

Another instance where the Organization met spectacular success was the eradiction of the rabbit menace. In 1950 it was estimated that there were about 600 million rabbits which were eating the best grass, enough to feed 100 million sheep. The introduction of a parasite from South America spread an epidemic which killed the rabbits in a matter of months. The mosquitoes acted as carriers of the disease in this case.

Another success achieved by the Organization was in the investigation of the causes of very poor soil in a particular region. The investigation showed a deficiency of zinc and copper and when this deficiency was made up the production increased 20 to 40 fold. Sir Ian also referred to the changing face of the country in Southern Australia where the addition of 2 ounces of molyb-

denum to an acre was sustaining much plant growth, and lakes and oceans of green were springing up in the country.

He mentioned the experiments on the cross-breeding of Sindhi and Australian cattle. In the European cattle introduced in Australia earlier, sweat does not evaporate easily whereas the Pakistani cattle loses heat more freely. In the Australian climate, he said, the latter characteristic is an advantage.

Symposium on Science and Industry

The Pakistan Association of Scientists held a symposium on "Science and Industry" on January 20, 1958 at the Y.M.C.A. Hall, Karachi. Mr. Muzaffar Ali Khan Qizilbash, the then Central Minister for Commerce and Industries, inaugurated the symposium. Mr. Ghulam Faruque, Chairman, P.I.D.C., Dr. Salimuzzaman Siddiqui, Director, Scientific and Industrial Research, Mr. Hatim A. Alvi, Prof. M.A.H. Qadri of the Karachi University, Mr. M.A. Azam of the Industrial Survey Organization, and Mr. Kasim Dada took part in the discussion.

In his inaugural address, Mr. Qizilbash emphasized the importance of advancement of science and research in raising the standard of living of the common man, and urged the philanthropists and industrialists of the country to make their contributions towards this end. He said that even for sheer self-interest it is an investment which pays rich dividends in many ways. The Government, he said, had set up various research institutes and testing laboratories in both the wings of the country.

Mr. Ghulam Faruque, who presided over the conference, gave a brief history of the P.I.D.C. In the socialist countries, he said, the governments are taking over industrial and commercial establishments, whereas in Pakistan the Government is reversing this trend by setting up factories through the P.I.D.C. and then handing them over to private concerns.

Mr. Faruque referred to cheap power, cheap cement and cheap fertilizers as the three essentials to improve the economy of the country.

Addressing the symposium, Dr. Siddiqui gave a brief account of scientific and industrial research in the country and dealt with some of the important problems. He referred to Rauwolfia, which is being considered as the drug of choice in the treatment of high blood pressure and mental ailments. To put a new drug on its feet, he said, needed a colossal amount of collective work and it was only with such co-operative research work in America and Europe that Rauwolfia was coming into its own as one of the most important medical discoveries of the recent times. He said that unless we follow up our investigations on drugs in an integrated manner the ultimate credit of new findings will slip away from our hands.

Mr. Hatim Alvi said that the private capital must play its effective role in the field of scientific research. If research in the industrial field was not carried on, he said, it will not be the scientists who will suffer, it will be the capitalists who will be wiped out.

Prof. M.A.H. Qadri, Mr. M.A. Azam and Mr. Kasim Dada also spoke on the importance of scientific research in the field of industry.

Pakistan Association of Scientists and Scientific Professions

The amalgamation of the Pakistan Association of Scientists and the Pakistan Association of Scientific Professions has been announced. The new association has been named as the Pakistan Association of Scientists and Scientific Professions.

Applications Filed in the Patent Office

The total number of applications for patents and designs filed in the Pakistan Patent Office during the years 1948-1957 is given in the following table :—

	Patents		Designs	
Year	Applications	Accepted	Applications	Registered
1948	734	nil	6	nil
1949	1089	204	38	2
1950	695	831	54	43
1951	733	1104	85	38
1952	717	713	106	66
1953	761	573	55	36
1954	959	505	81	68
1955	908	452	65	35
1956	904	1052	114	70
1957	1024	213	255	327

Canada-Pakistan Accord on Patents

A convention between Canada and Pakistan concerning the priority of inventions was signed in Karachi on the 15th January, 1958. The convention provides generally that national treatment shall be reciprocally granted to citizens of the other contracting party and to bodies incorporated or registered under its appropriate legislation. Thus a person who has first made application for a patent in Pakistan shall have the right, within twelve months from the date of the application, to claim priority in Canada for an application in respect of the same invention. The convention shall come into force three months after signature.

Pakistan has established similar reciprocal arrangements with Australia, Ceylon, India, New Zealand, Union of South Africa and United Kingdom.

Forthcoming International Conferences

International Mathematical Union, 3rd General Assembly ; Aug. 11-13, 1958 ; St. Andrews, Scotland.

International Astronomical Union, 10th General Assembly ; Aug. 13-20, 1958 ; Moscow, USSR.

International Congress of Mathematicians ; Aug. 14-21, 1958 ; Edinburgh, Scotland.

8th Quinquennial Congress of the Universities of the Commonwealth (Association of Universities of the British Commonwealth) ; Sept. 1-5, 1958 ; Montreal, Canada.

Peaceful Uses of Atomic Energy, Second International Conference ; Sept. 1-13, 1958 ; Geneva, Switzerland.

3rd Session of the Congress of the World Meteorological Organization ; Sept. 3-23, 1958 ; Geneva, Switzerland.

6th International Congress of Tropical Medicine and Malaria ; Sept. 5-13, 1958 ; Lisbon, Portugal.

International Pharmaceutical Federation, 17th General Assembly, and 18th International Congress of Pharmaceutical Sciences ; Sept. 8-15, 1958 ; Brussels, Belgium.

6th General Assembly and 7th Technical Meeting of the International Union for the Conservation of Nature and Natural Resources ; Sept. 11-19, 1958 ; Athens, Greece.

Symposium on Solar Heating (Association for Applied Solar Energy) ; Sept. 22, 1958 ; Phoenix, Arizona, U.S.A.

International Conference on Scientific Information (Sponsored by American Documentation Institute, National Academy of Sciences—National Research Council, and the National Science Foundation) ; Nov. 16-23, 1958 ; Washington D.C., U.S.A.

BOOK NOTICE

L' Age Nucleaire

A specimen copy of the above journal (Issue No. 8, Jan./Feb. 1958) has been received. The journal is publised two-monthly, in French, by the Compagnie Francaise d'Editions, 40, Rue du Colisee, Paris 8; the subscription rate abroad is 4000 francs per annum. The field covered is indicated by the following translated list of contents of this issue:—

R. Gibrat: Swimming-pool reactors; D. A. Bell: Nuclear power stations in the national economy; M. Bayet: The concept of "effective section"; P. Baruch: The Physics of semi-conductors; D. Barbier: The chemical analysis of the atmosphere, of the sun and of the stars; R. Henry and S. Davydoff: The International Conference on the use of radioisotopes in scientific research; D. Blanc: Nuclear science course (Structure and stability of the atom); J. Jarry: Further discussion on the satellite problem; J. Delobeau: Scientific comments on the Russian satellites; D. Blanc: Particle accelerators and detectors; W. Pilz: The Astronautical Congress in Barcelona; M. Rolland: Electronics.

PATENT NEWS

Abridgements of patent specifications of Pakistani inventors notified as accepted during 1957 in the Part IV of the Gazette of Pakistan

106508 *A strain gauge for measuring the strength of a wire.*—M.M. HASSAN KHAN. The gauge consists of three rotatable pulleys, two of which are rigidly mounted on a rigid frame and the third pulley which is intermediate the said two pulleys are resiliently mounted and is slidable in relation to the said frame, and a pointer fixed along the said slidable pulley to travel along a scale.

106713 *A mechanism for automatically tilting the frame of the chimney of a hurricane lantern.*—SIND INDUSTRIAL CORPORATION. The mechanism comprises an inverted L-shaped extension of the lantern's handle which when lowered presses steel notches fixed in the top frame of the chimney upwards, thereby raising the upper frame of the chimney. The upward movement is transmitted to the lower frame through two steel wires bent at both ends in the form of hooks, which are hinged in the notches of the upper frame. As the lower frame is hinged, only the side opposite is raised, enabling the chimney to tilt. The frame in its tilted position is supported by a steel spring. The wick is then lighted and the handle is raised making the chimney to revert to its former position.

106813 *A solar cooker.*—ZAFAR AHMAD. The cooker comprises a conical reflector lined at sides and the base with thin silvered glass sheets, carrying at its bottom an adjustable bracket for placing cooking utensils and one transparent glass or plastic cover held centrally to close the utensils. The glass cover above the utensils, being diathermous, allows the rays to pass through and shields the utensils from the cooling effects of air currents.

107106 *An electrical device for preventing head-on collision of railway trains.*—S.M. AYUB. A metallic concave mirror having a convex lens is fitted exactly at the centre of every engine. Behind the centre of the mirror is housed a sensitive photoelectric cell which energises the magnet and completes the circuit which motivates an electric motor controlling a valve fitted in the stream feeding pipe so that steam is cut off. The circuit also motivates a timing device completing another electrical circuit which in turn operates upon the vacuum brakes of the train thus bringing it to a stand-still.

107180 *A solar water lift.*—C.A. AZIZ. The lift is characterised by a double walled solar boiler using low boiling point working substance which on vapourisation exerts pressure on the driving mechanism to lift water.

107390 *A damp-proofing powder.*—I.A. SHEIKH. The powder is prepared by causing hydrated lime to react first with a compound of caustic soda and mustard oil and then with alum solution.

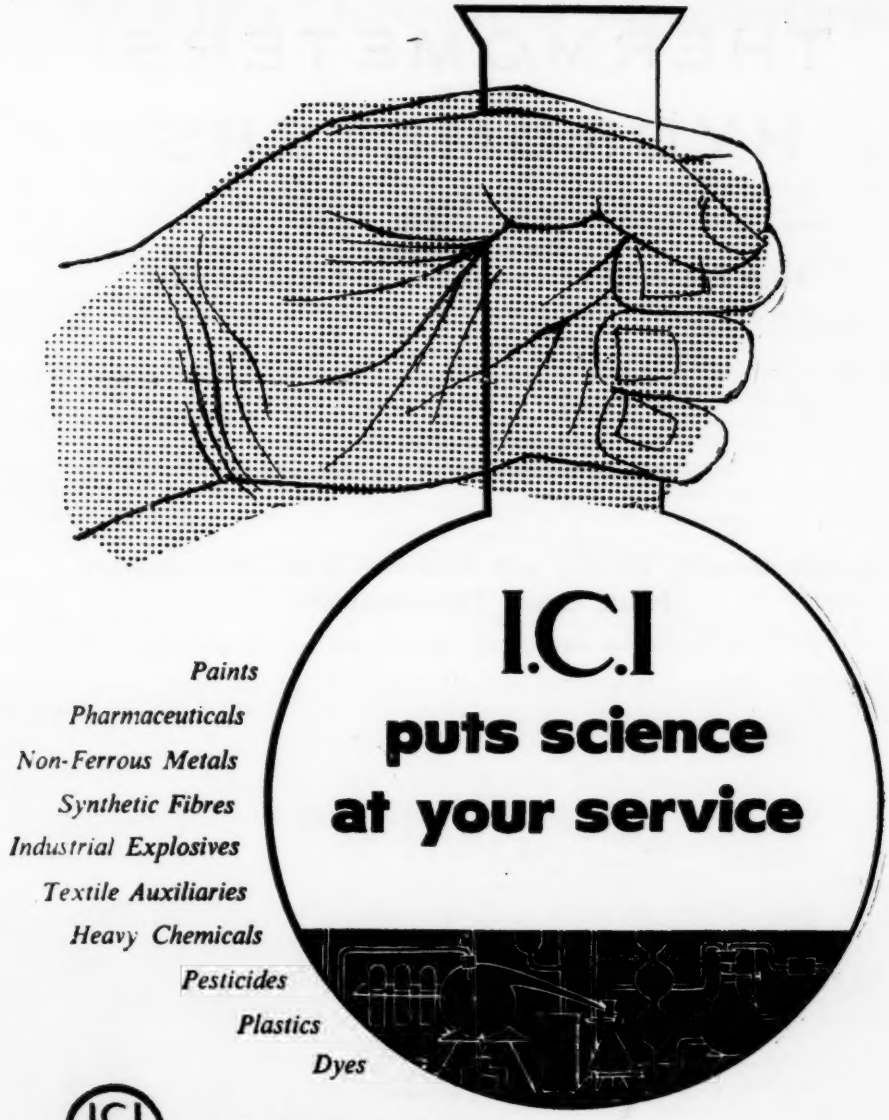
107393 *Liquid fuel oil burning apparatus.*—M.M. HANEEF. Steam is generated in a metal pipe and is carried to the orifice of the burner from where it is emitted into the stove taking with it liquid fuel oil. The mixture of steam and oil catches fire in stove and, striking against the flash wall of the stove, is diverted back under a heat resisting detachable steel plate placed on top of the metal pipe, to keep proper temperature for ignition and help the flame to burn for a longer period.

107505 *A petrol gas plant.*—A.H. CHOTANI AND S.M. ISHAQ ALI (of Pakistan Council of Scientific and Industrial Research). The gas plant comprises a gas accumulator, a high speed blower, a device for bubbling air through petrol contained in a tank, and means for leading the mixture of atomized petrol and air generated in the tank to the gas accumulator. The device for bubbling air through petrol is preferably made of a perforated hollow chamber kept submerged in the petrol.

107506 *A table cigarette box.*—A. RAHMAN. The box contains chambers for cigarettes and has a shutter device provided at one end for dispensing cigarettes.

107520 *Manufacture of light weight concrete.*—M.M. QURASHI, A.H. CHOTANI, A.K.M. AHSANULLAH AND M.A. CHOUDHARI (of Pakistan Council of Scientific and Industrial Research). The concrete is made by combining cement with rice husk ash preferably in a proportion of 1:1 by volume.

107561 *Manufacture of matboard.*—S.A. JUMANI. A mat made of bamboo cane and or palm leaves is glued to plywood and pressed.



I.C.I

puts science at your service

Paints
Pharmaceuticals
Non-Ferrous Metals
Synthetic Fibres
Industrial Explosives
Textile Auxiliaries
Heavy Chemicals
Pesticides
Plastics
Dyes

ICI *symbol of Quality and Service*

IMPERIAL CHEMICAL INDUSTRIES (Pakistan) LTD.
(A subsidiary of Imperial Chemical Industries Ltd., London)
KARACHI LAHORE CHITTAGONG DACCA

THERMOMETERS AND HYDROMETERS

EST. **ZECOL** 1888

High Precision Thermometers and Hydrometers

Guaranteed accurate and produced to the following specifications:—
Institute of Petroleum (I. P.)
American Soc. for Testing Materials (A. S. T. M.)
British Standards Institution (B. S. I.)
Standardisation of Tar Products (S. T. P. C.)

All sizes and ranges of

"Engraved on Stem" Thermometers

Spirit and Mercury filled

All sizes and ranges of

Specific Gravity, Density and Arbitrary Scale Hydrometers

Metal Cased Thermometers

For Oil and Petroleum testing and Industrial and Refinery purposes

G. H. ZEAL LTD.

REGD. TRADE MARK

Sole Agents in Pakistan for Laboratory and Industrial Thermometers.

MESSRS. HARDCASTLE, WAUD (PAKISTAN) LTD.

Telegrams: **"WAUDCOY"** *Telephone:* **31772**

42-44 Laxmi Building, Bunder Road, Karachi, Pakistan.

Branches: CHITTAGONG No. 1 Jail Road, (Tele 5733)
LAHORE No. 104 McLeod Road, (,, 5733)

Associated Instrument Manufacturers (Pakistan) Ltd.

Representing

SALES AGENTS IN PAKISTAN, FOR:

Laboratory Equipment for Education and Scientific Research

R. & J. BECK LTD.	Microscopes and Accessories.
CHANCE BROS. LTD.	Optical glass, Colour filters, Microscopes slides and Cover glasses, Veridia tubing.
EDUCATIONAL & SCIENTIFIC PLASTIC LTD.	Osteological models and demonstration units for medical education.
GRIFFIN & GEORGE LTD.	Comprehensive laboratory apparatus, instruments and furniture.
HILGER & WATTS LTD. (HILGER DIVISION)	Spectrographic equipment, Absorptiometers, Flourimeters, Polarimeters, Refractometers, X-Ray diffraction equipment etc., etc.
JOHNSON MATTHEY & CO. LTD.	Laboratory platinum ware.
JAMES SWIFT & SONS LTD.	Petrological and Metallurgical microscopes and accessories.
THE TINTOMETER LTD.	Lovibond Tintometers and Comparators.

Equipment for Survey & Meteorology

HILGER & WATTS LTD. (WATTS DIVISION)	Theodolites, Surveying and Engineer's Levels, Alidades, etc., etc.
C. F. CASELLA & CO. LTD.	Meteorological instruments and apparatus.
SHORT & MASON LTD.	Meteorological instruments.

Other Optical Instruments

BARR & STROUD LTD.	Binoculars and Rangefinders.
BROADHURST CLARKSON & CO. LTD.	Telescopes.

Industrial, Marine & Drawing Office Equipment

CAMBRIDGE INSTRUMENT CO. LTD.	Electrical process indicators and controllers etc.
EVERETT EDGECUMBE & CO. LTD.	Electrical indicating and recording instruments etc.
ISOTOPE DEVELOPMENTS LTD.	Nucleonic equipment for industry.
LONDEX LTD.	Automatic electrical control apparatus.
SMITHS INDUSTRIAL INSTRUMENTS LTD.	Tachometers, Chronocounters, Remote position indicators, etc.
CHANCE LONDEX LTD.	Marine electric lights and Buoys.
GLENIFFER ENGINES LTD.	Marine diesel engines.
STONE CHANCE LTD.	Marine and aerial navigation lighting equipment.
E. N. MASON & SONS LTD.	Photo-copying, plan printing machines.
A. G. THORNTON LTD.	Drawing instruments, slide rules etc.

Medical, Surgical & Optical Supplies

COX-CAVENDISH ELECTRICAL CO. LTD.	Electromedical equipment etc.
GOWLLANDS LTD.	Diagnostic instruments.
THE HADLEY CO. LTD.	Spectacle frames.
THE UNITED KINGDOM OPTICAL CO. (BAUCH & LOMB) LTD.	Spectacle lenses and blanks.

Registered Office

3rd Floor, Mohammadi House
McLeod Road, Karachi
Phone : 34326

WE CAN SUPPLY THE FOLLOWING SCIENTIFIC INSTRUMENTS FROM OUR FOREIGN PRINCIPALS:

Spectrophotometers, Goniometers, Powder Cameras, Colorimeters, Absorptiometers, Flame Photometers.

Physical Apparatus for teaching purposes.

Rotary Pumps, Oil Diffusion Pumps, Compression Vacuum Gauges, Mechanical Boosters, Leak Detectors, Electron Microscopes, High Vacuum Pumps, Mercury Ejectors, Atom Models for Schools, Electrical Measuring Instruments, Polarographs.

Transistors.

pH Meters, Galvanometers, Polarometers, Densitometers, Potentiometers, Thermo-Electric Thermometers, Measuring Amplifiers, Radiation Balance Meters.

Condensers, Crucibles, Dessicators, Extraction Apparatus, Filter Paper and Tubes, Gas Generators, Centrifuges, Furnaces, Hot Plates, Incubators, Microscopes and Accessories, Mills, Molecular Weight Apparatus, Pyrometers, Refractometers, Thermoregulators, Viscometers, Wasserman Apparatus, Microtomes, Sterilising Equipment, Vaccine Equipment, Oil Testing Apparatus, Coal and Coke Testing Apparatus, Gas Analysis Apparatus, Metallurgical Apparatus.

All types of Laboratory Glassware.

INTERNATIONAL INDUSTRIES LTD.

HAKIMSONS BUILDING, 1, WEST WHARF ROAD, KARACHI

ALL

MACHINERY & EQUIPMENT

FOR THE

PHARMACEUTICAL, CHEMICAL AND COSMETIC INDUSTRIES

ALSO SMALL-SCALE PRODUCTION

INSTRUMENTS & APPARATUS FOR LABORATORIES

AND CHEMIST SHOPS

Please communicate your needs to:

JOSEF DECKELMANN

P. O. BOX 766, ASCHAFFENBURG/WEST GERMANY

WITHSTANDS All WEATHER

Rains, heat and dust, cause heavy damage. See that Your goods are well covered and fully protected with our

100% Weatherproof

TARPAULINS

Famous for Reliabiliiy & Quality

MODERN CHEMICAL & TARPAULIN INDUSTRIES

Office Tele : 37385

Ameejee Valeejee Building,
Frere Road, Campbell Street,
Karachi.

Shop Tele : 32973

A/6 Ketchi Gali No. 3,
Para Line, Near Jodia Bazar,.
Karachi.

KILBURN & CO. (PAKISTAN) LTD.

MANESTY MACHINES LTD.
- Tablet Making Machines
- Water Stills
- Drying Ovens
- Mixers, Granulators, etc.

CLIFFORD PACKAGING MACHINERY LTD.
- "Newman" Gumming Machines
- "Newman" Labelling Machines.

ALBRO FILLERS & ENGINEERING CO. LTD.
- Liquid & Powder Filling Machines.

AUTOPACK LTD.
- "Ampoule" Washing Machines
- "Ampoule" Filling & Sealing Machines
- "Ampoule" Cutting Machines

BRAMIGK & CO. LTD.
- Pulverisers & Mikro-Atomisers.

Agents :

KILBURN & CO. (PAKISTAN) LTD.

AKMO BUILDING, 1, WEST WHARF ROAD, KARACHI.
Telephone : 32537

29/1, TOPEKHANA ROAD,
P. O. RAMNA, DACCA.
Telephone DACCA 3262

AGRABAD ROAD,
CHITTAGONG.
Telephone : 5961

48-2-66 KONTAKTS

Here with a Loaf of Bread beneath the Bough,
A Flask of Wine, a Book of Verse - and Thou
Beside me singing in the Wilderness -
And wilderness is Paradise enow.

'OMAR KHAYYAM'

In these modern days we still regard with some envy the happy aspect of Omar Khayyam's philosophy but engineers know that more than dreams are necessary to keep the wheels of industry moving with maximum efficiency.

It's part of Burmah-Shell's service to study all lubrication problems in order to minimise maintenance cost without running the risk of damaging valuable machinery.

That is why factory managers all over Pakistan are calling in Burmah-Shell Technical Service to help them sort out tricky lubrication problems. Their advice is freely given, and their planning reduces maintenance cost.

BURMAH-SHELL *is part of progress in Pakistan*

BSP-246

Printed at
Nazeer Printing Works
Water Course Road, Karachi